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CHANGES IN BLOOD IN CONNECTION WITH THROMBO-EMBOLISM

AN INVESTIGATION REGARDING OPERATION AND DELIVERY

BY

GUNNAR BERGQUIST

STOCKHOLM 1945

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FROM THE STATE INSTITUTE OF HUMAN GENETICS AND
RACE BIOLOGY, UPPSALA
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Preface

This work presents the results of investigations into a number of blood changes after operation and childbirth, as also investigations into a small number of cases of thrombosis arising in conditions of this kind. Because of the increased risk of thrombosis after operation and childbirth it seems probable that, if changes in the composition of the blood influenced the onset of thrombosis, these events would be generally followed by such changes. Furthermore, such changes might be expected to be especially strong in thrombosis cases. The problems bearing on the genesis of thrombosis are complicated; to provide a background to the investigations, this work begins with a comparatively lengthy survey of the literature. The factors subsequently investigated in the book are primarily the adhesion tendency of the leukocytes, the number of blood mast cells, the coagulation time of the blood, and the thrombin effect. The material of operation cases comprises 445 individuals, and the childbirth material 110 individuals. Besides these, 39 cases of thrombosis have been investigated.

Dr Erik Waller, M. D., Stockholm, under whom I have previously worked, awakened my interest in investigations of this type, and placed his material at the Surgical Department of the Lidköping hospital at my disposal; for all this I beg to thank him most warmly.

Material has also been obtained from the Surgical Department of *Karolinska sjukhuset*, Stockholm, the Surgical and the Obstetric Departments of *Akademiska sjukhuset*, Uppsala, from the Surgical Departments of the Västerås and the Örebro hospitals, and from the hospitals at Köping and Arboga. I should like to offer my hearty thanks to the Head Physicians whose kind help enabled me to obtain material; these are Professor John Hellström, Professor Olle Hultén, Dr Lennart Norrlin M. D., Dr Gösta Bohmanson M. D., Dr Gustav Levander M. D. and Dr Bertil Lindstrand.

The material was subsequently analyzed at the State Institute of

Human Genetics and Race Biology, Uppsala, and I beg to tender my cordial thanks to Professor Gunnar Dahlberg for the working facilities he afforded me, and for advice and directions while the work was in progress. I am also indebted to Professor Robin Fåhræus for his kindness in allowing me to consult with him on certain important questions. In this connection I should like to offer my warm thanks to Dr Erik Jorpes M. D., who has given me valuable advice and suggestions. Docent Hjalmar Holmgren has also been most generous with valuable information on the methods of determining the number of mast cells.

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Arboga, April 1945.

G. Bergquist.

CHAPTER I

Survey and Discussion of Earlier Investigations

Not until the end of the 18th and beginning of the 19th centuries are more definite observations bearing on thrombosis to be met with in the literature. Virchow's works (1846—1856) introduced more rational viewpoints upon the question of the occurence of thrombosis. Virchow maintained (1854) that thrombosis is due to a combination of causes, one of which being local inflammation in the vascular wall. He furthermore stressed the increased tendency of the blood to coagulate, and, in fact, he considered the thrombi to be nothing else but clots. Clotting may be induced by mechanical factors, a changed flow of the blood in general, or local circulatory weakness.

Virchow clarified an important circumstance in demonstrating the causal connection between thrombosis and embolism. His view that thrombosis was a local process was thereby strengthened, and the foundation laid for his theory that the underlying cause was a coagulation of the blood arising mechanically (by stagnation of the blood), or chemically (because of alterations in the chemical composition of the blood).

Further light was thrown on the problem by investigations into the histological structure of the thrombi. As has been indicated above, Virchow considered that the thrombus was only a clot, and this view was the accepted one until Mantegazza (1869) and Zahn (1881) observed direct under the microscope how fibrin-free thrombi were formed by leukocytes and some granular material. These investigators saw a fundamental contrast between these fibrin-free thrombi and the red coagulation thrombi, and called the former 'white thrombi'. An important point is that this observation showed that a thrombus does not need to contain all the constituents of the blood; certain definite corpuscular elements may separate off in the flowing blood and agglutinate, forming solid conglomerations. This

indicates that certain conditions in the blood stream are necessary for the genesis of thrombi, a fact which v. Recklinghausen (1883) points out. Bizzozero (1882) showed that the above-mentioned granular formations consist of thrombocytes, thereby completing the identification of all the structural elements of the white thrombus.

Eberth and Schimmelbusch (1888) saw that, when the blood stream was slow, these thrombocytes made their way out towards the periphery, adhering to the wall if the vascular endothelium was injured. When new thrombocytes were added, together with leukocytes, the white thrombus was gradually formed and could finally close the lumen altogether. The coagulation of fibrinogen following this formation of a white thrombus was apparently caused by ferments given off from the agglomerated blood cells. Thus, the mechanical factor dominates the minds of these investigators, also, even though they maintain that irregularities or lesions in the vascular wall may also dispose to thrombi.

Aschoff and his co-workers have made experiments into the significance of different kinds of irregularities in the vascular wall. They investigated flowing suspensions, made to pass special transverse and oblique obstacles, with a view to discovering by experiment how far deposition of particles takes place. These experiments will be discussed in more detail later; all that need be said here is that experiments made in this way strengthen the old view that lesions in the vascular wall are of importance for the occurrence of thrombi, in which, however, mechanical factors also play a material part (Aschoff, de la Camp, Beck, Krönig, 1912).

Virchow, too, discusses the importance of the vascular wall for thrombus formation. As has been indicated, he considered local inflammation in the wall to be one of the aetiological factors, and chemical changes in the blood to exert influence. Baumgarten (1877) verified Hunter's (1784) earlier observation that the blood in a doubly ligated vein may remain in a fluid state for a long time. If, however, the vascular wall was injured and became infected, the blood coagulated. As a result of this investigation, the view that endothelial lesion and inflammation (in the vascular wall) are of primary importance came to dominate in the discussion round this problem. French authors in particular took up the enquiry and stressed this causal connection (Vaquez, 1894).

The view that a change in the composition of the blood, i. e. its

chemical or morphological constituents, played a part in the appearance of thrombosis had, as already stated, been advanced fairly early on. Nevertheless there was no clarity as to this question until the beginning of the 20th century, due in part to purely technical difficulties. The methods of analysis were uncertain and faulty, and present-day methods of investigation were then unknown. It had long been known that the composition of the blood could undergo changes in connection with different diseased states, and it was natural that such changes should be placed in relation to the occurrence of thrombosis.

The above brief review of the older literature on the subject shows that Virchow's triad—1) changes in the blood stream, 2) lesions in the vascular wall and 3) changes in the composition of the blood —covers the view then prevailing. This view has stood the subsequent test of time fairly well. One or other of these three factors may, at different periods, have dominated the discussion of the topic, but the basic theory has on the whole remained unchanged.

Mechanical Factors

In principle, both chemical and mechanical factors may be thought to exert influence in the occurrence of thrombi. If chemical factors alone decided the issue, and differences in the blood stream had no importance, we would have to reckon with a general coagulation of the blood—provided the chemical change extended over the entire amount of blood. A local precipitation of substances would then have to be attributed to a chemical change proceeding from a point or a region on the vascular wall which only affected the blood passing by at the moment. Even were this so, it would nevertheless be reasonable to assume that the speed at which the blood passes is of importance.

In point of fact it is *a priori* probable that mechanical factors exert a material influence. Attempts to determine this must pay regard to 1) the speed of the blood stream and 2) the relation of the blood stream to the vascular wall. Regarding 2) we must know a) the effect of irregularities in the wall and b) the effect of the vessel's calibre.

Obviously, thrombi will form more readily if the blood stream is

slow than if it is rapid. And indeed, various investigators support the view that the retarded passage of the blood through the vessels is the main cause behind the formation of thrombi, changes in the composition of the blood or in the vascular wall being only contributory factors. As has already been indicated, Zahn, v. Recklinghausen, Eberth and Schimmelbusch have shown by experimental studies that the lowered rate of the blood stream should be assigned an important part in thrombosis; they maintain, moreover, that this decrease in speed is to be looked upon as the main factor.

Aschoff (1892, 1912 a, 1912 b) asserts that thrombosis in the veins, i. e. the progressive, spontaneous thrombi which the present author has in mind here and in the following pages, depend both for their origin and their structure on the rate of the blood stream, above all on the type of stream. To begin with, he demonstrates that investigation of a thrombus shows it to be made up of three different sections: 1) the head part, or the white thrombus, 2) the neck part, or the mixed thrombus and 3) the tail part, or the red thrombus. In a histological examination of these different sections it is possible to differentiate in the head part a peculiar structure of parallel lamellae or 'beams' and transverse beams, so arranged that the whole resembles a colony of corals. The beams are made up of aggregated thrombocytes, and there are leukocytes along their edges and in the interstices. Supported by these observations, Aschoff states that the explanation of this strange structure must lie in something other than lowered rate of flow. By studying artificial streams in a vascular system in whose walls both straight and obliquely placed obstacles had been introduced, he tried to clarify the effect of such obstacles on a suspension flowing through the system. He used sawdust silted up in water, and showed that this was deposited in different places in the vessel, due to changes in the type of the flow, which was determined by the above-mentioned obstacles. The cause of this variation in the placing of the deposits could be observed direct: the flowing suspension was seen to form vortices of different types at the different obstacles. One type was found at the straight barriers, another at the transverse ones, recalling vein valves, and yet another at the places where a thin vessel opened into a thicker one. The structure of the deposit also depended on the type of vortex. Thus, it could be seen how, to start with, undulating or net-like deposits were formed in the rim of the vortices. On

the strength of these observations, Aschoff asserts that the lamellar structure of the thrombus arises primarily in this manner, and states at the same time that the structure cannot be due to a secondary reconstruction. He also shows that the same deposit is obtained on an irregular surface as on a smooth one. Aschoff demonstrates furthermore that the lamellar structure in the deposit appeared only in tests with flowing suspensions, and that deposits of this kind do not appear in still water which has been slightly disturbed. As, however, he neither gives any details as to the character of the suspensions he uses (only saying that he takes sawdust) nor states whether the stream is flowing in an open or a closed vessel, it is difficult to determine what support he has for his pronouncements about the deposition. The suspensions must, however, be heterogeneous if different layers resembling those in the thrombus are to arise.

Aschoff's views in this question were not adopted by all investigators. A different opinion was advanced by Ribbert (1912), who claimed that it was the irregularities (lesions) in the vascular wall that constituted the main cause of the thrombosis. When a formation of this kind occurs, thrombocytes and, later on, leukocytes stick fast, producing the first foundation of the thrombus, which is subsequently built upon. He ascribes importance to a lowered rate of blood flow only inasmuch as it is less likely to dislodge already adhering particles. In support of his view, Ribbert maintains that the obstacles inserted by Aschoff can in no way be compared with the vein valves. These latter do not consist of rigid processes but are mobile; sometimes they lie close along the vascular wall, sometimes they are right out in the blood stream, and then, too, they are in constant motion. He further maintains that the quantity of the different kinds of 'beams' and lamellae present in a thrombus would call for an enormous number of different vortices, which must in addition be constant during some period. He also points out that the oldest part of the thrombus, the part which according to Aschoff is mainly caused by vortex formations, presents least lamellar structure. After these speculative reservations, Ribbert describes a number of experiments, which he considers prove the accuracy of his view. He cauterized with lapis the exposed jugular vein of rabbit and extirpated the vessel after 3– 10 minutes; this he hardened and cut longitudinally. The injured intima showed polypous formations

of thrombocytes with irregular, secondary deposits of leukocytes. According to Ribbert, these thrombi cannot be due to vortex formations. The sections also showed how, on account of the cauterization, the blood had been injured and adhered to the vascular wall in small 'icicles', somewhat reminiscent of the obstacles in the stream described by Aschoff. If the vortex were the cause of the thrombus, then thrombosis should have been observed behind these 'icicles'. This is not so: the thrombus is formed on the icicle's tip. Ribbert has also drawn a thread through the blood vessel and showed that small thrombi, formed of thrombocytes, adhere very rapidly to it. He maintains that if the thread is to be regarded as an obstacle analogous to the one used by Aschoff, the thrombus should confine itself wholly to the downstream side of the thread. This is not the case, however: the thrombus is built up round the thread. Its first layer is not here a porous arrangement of beams; it is homogeneous and solid, and recalls the structure which the oldest part of the spontaneous thrombus presents in man. Finally. Ribbert maintains the following: if vortices really had the importanc eascribed to them in this connection they would divide up the three form elements of the blood in such a way that the thrombocytes would first be made to flow in separate paths, into which the leukocytes would then be forced, while the erythrocytes flowed on. He also points out that there is only one constituent in the sawdust suspensions used by Aschoff, whereas the blood contains three, with different specific gravity. Ribbert goes on to say that it is inconceivable that the blood platelets everywhere present among the erythrocytes should not take these latter with them, instead of forcing their way out between them to collect at appointed places. To support his view, Ribbert made the following experiment. He drew a thread through the jugular vein (of rabbit) and cauterized the vessel at some distance thence with lapis. Immediately after this he injected a suspension of cinnabar into the ear vein on the same side. The thrombi formed on the thread and on the intima injured by cauterization were found, on investigation, to be fairly uniformly interspersed with cinnabar granules, which had another specific gravity than the thrombocytes. Ribbert considered this to prove that it is not vortices but lesions in the wall which constitute the primary factor in thrombosis.

Ribbert's work met with opposition from Beneke (1913) who declared himself unshaken in his adherence to Aschoff's theory as

being the correct one—without, however, contesting Ribbert's pronouncements in any detail.

It may be mentioned here that, in artificial experiments, an obstacle for the stream may be produced in the form of a ridge on the wall itself. This ridge then has the same smooth surface as the rest of the wall in the tube. Obstacles of this kind would hardly arise in a blood vessel, where such a ridge would as a rule probably be due to a vascular lesion, the wall's surface then also becoming more or less irregular. On the other hand, normal vessels do show equivalents to the artificial ridges from experiments in so far as an increased friction against the stream is set up in sharp bends and where branches are given off. Vortices can arise here. If thrombi have a predilection for such places, it is because a retarded blood stream combined with a purely mechanical factor predispose favourably to thrombosis. If, on the other hand, a surface shows irregularity, it ought to be possible for a thrombus to occur even in a vessel which is straight and unramified. The circumstance established by several pathologists, e. g. Aschoff, that thrombi tend particularly to arise at places where vessels divide is most readily interpreted as indicating the importance of an isolated mechanical factor. Naturally, this does not exclude the possibility of influence from irregularities. The question here is not necessarily one of alternatives; it may be one of combination.

Aschoff's view that retardation of the stream is the main cause of thrombosis has several adherents among investigators, although they do not advance any new grounds for their opinion. Of these we quote Vietor (1925), v. Jaschke (1926—27), Öwre (1929), Walters (1930), Dahl-Iversen and Ramberg (1932).

The previous pages have discussed the decrease of the rate of the blood stream as cause of thrombi, without taking into account the different factors which may be thought to retard the stream. In itself any discussion of these factors is of no very great consequence. Nevertheless, it is interesting to be able to see that thrombi arise primarily in persons suffering from diseases lowering the rate at which the blood flows in the vascular system, or in those where the rate has been lowered for some other reason. This being so, the different factors which may affect the rate of flow will be touched on in connection with the works of a number of investigators who have interested themselves in this question. The factors in question are

1) cardiac and vascular diseases, 2) stasis in the veins of the lower parts of the body, due to mechanical conditions, 3) operation and 4) static states (recumbency).

The importance of the *cardiac diseases* was early on maintained by several authors, but only with present-day major statistics of autopsy cases has it been possible to verify the observation from the sick-bed, long known in practice, that cardiac insufficiency (in operated and post partum cases) often sets up thrombosis. Hermansson (1925) submits a material in which 18 of 26 cases with embolism showed cardiac lesion to a greater or lesser degree. Walters (1927) states that thrombosis very often occurs in cardio-vascular diseases. He describes investigations made by him which show that the blood pressure always falls after operations, indicating circulatory disturbance. Schumacher (1926—1927) describes a material in which 53 % of the thrombosis cases show lesions in the circulatory system, while the figure for the embolism cases is 70 %. Fahr (1927) collected thrombo-embolism cases on which autopsies had been performed during 1925—1927, and showed that cardiac or vascular disease occurred in 54 of 95 cases. Höring (1928) and Oberndorfer (1928) also submit on the whole similar figures. Schleussing (1929) found that, of 27 cases of fatal embolism, all showed cardiac or vascular lesions of more or less serious character. Sulger (1931) made collective statistics of cases communicated by a number of authors, and calculated them according to statistical methods: he found the theory as to the importance of cardiac and vascular diseases to be well founded, for he sums up: '. . . dass in der überwiegenden Mehrzahl der an Thrombosen und Embolien Gestorbenen, Erkrankung des Herzens und der Gefässe nachzuweisen sind'. Of later publications we can mention one by Culp (1940) in which he states that lowered blood pressure may in certain cases be of importance for thrombosis. Culp furthermore stresses the fact that, in the material he investigated, a third of the cases complicated by thrombo-embolism had cardiac insufficiency before operation.

The great importance that the cardiac and vascular diseases have for the occurrence of thrombosis is also clearly illustrated in the following table, taken from R. Geissendörfer (1935).

Table 1

Author	No. of fatal lung embolisms	Cardiac and vascular diseases
C. Bauer	—	95 % of the cases with non-normal heart.
Clairmont	—	70 % yielding pathological heart findings at autopsy.
Fahr	95	54, i. e. 57 ± 5.1 % cardiac and vascular disease.
Fritzsche	16	9 cases with diseased heart, of which 6 not discovered until autopsy. Of 19 cases of healed lung embolism, only 3 with clinical heart findings.
Höring	65 (med.)	50 cases with cardiac change, weakened heart or several lesions together (i. e. 76.9 ± 5.2 %).
	51 post op.	32 cases with cardiac change, weakened heart or several lesions together (i. e. 62.7 ± 6.8 %).
Kazda and Stöhr	79 post op.	43 cases of cardiac and vascular changes at autopsy (i. e. 54.4 ± 5.6 %).
	68	49 cases of cardiac and vascular changes at autopsy (i. e. 72.1 ± 5.4 %).
Killian	58	35.5 ± 6.2 % with circulatory diseases (44 post op.), 67 ± 6.1 % fatal lung embolisms among the hopeless cases.
Kuhn	105	70 ± 4.4 cardiac and circulatory diseases.
Martini and Opitz	—	61 % cardiac disease.
Oberndorfer	97	2/3 of all cases of diseases of the circulatory system.
de Quervain	249 (not only fatal cases)	235 cases with concurrent diseases; 144 cases with cardiac and circulatory diseases; 53 cases of pulmonary and 11 of renal diseases; 2 cases of diabetes; 2 cases of adipositas; 1 case of alcoholism.
Rupp	56 post op. 601 (med.)	39 cardiac, 22 renal and 16 severe vascular diseases; 426 cardiac, 537 renal and 280 severe vascular diseases; 170 pulmonary complications.
Schumacher	—	50 % cardiac diseases.
Schleussing	—	100 % cardiac and circulatory diseases.

Several present-day investigators consider delayed emptying of the venous system of the lower parts of the body to be responsible for the reduced speed of the blood stream, and consequently to be the cause of thrombosis, also. Several of them claim to hold this opinion, but give no real reasons for doing so. Examples of these are Vietor (1925), de Courcy (1929), Aschoff (1934) and Geissendörfer (1935). Lindsay (1925) considers that stasis in the veins of the legs and the pelvis must be the main cause of thrombosis. In support of this view he states that thrombi are more often localized in the left leg than in the right. This he connects up with the situation of the sigmoid, inasmuch as the sigmoid and the rectum are so placed as to exert, when filled, a stronger pressure on the vessels. and thereby produce stasis. He sees another factor in the less direct drainage constituted by the left iliac vein. However, an inspection of Lindsay's figures shows that the material (40 cases of thrombosis in the veins of the pelvis and legs, of which 20 were in the left leg) is obviously too small to allow of such conclusions. Moreover, Schulte (1932) shows in a sufficiently large number of cases that thrombosis has no greater predilection for the veins of the left leg than for those of the right.

In his work of 1934, Robertson subscribes to the view that thromboses are due to stasis or stagnation of the blood. He states that aneurysms are often filled with thrombus masses. He goes on to state that, in heart diseases, thrombi may form in the auricles. Finally, he points out the usual autopsy findings of phlebolites in the venous plexus of the prostate. Basing himself on these circumstances, he makes the following remarks: 'It is surely neither injury to the vascular wall, nor infection, nor increase in the coagulability of the blood. It is simply stasis or stagnation of the circulating blood at these particular situations.'

Purely static factors are submitted by many investigators as being the main cause of retardation of the blood stream and onset of thrombosis. Thus, Aschoff maintains in his works that recumbency makes for changed conditions in the flow of the blood, particularly in the legs, due to diminished muscular work. In this connection it may, however, be recalled that the blood flows away more readily with the limbs in the horizontal position than when they are in the upright position. Without elaborating the reasons for his view, Walters (1927) maintains that investigations have shown the mere lying in bed to

make for impaired circulation in the veins of the legs, and that this fact should be ascribed importance in thrombosis. In a method worked out by himself, Frimann-Dahl (1935) has gone into the rate of flow in venous blood under different conditions. One of the things he has shown is that the flow in the veins of the legs becomes appreciably less rapid during confinement to bed. When, in addition, there is an operation, there is a far greater retardation of the blood stream, which may even almost stagnate. Two patients with a slow stream developed thrombosis; on the other hand, others with an equally slow stream showed no discernible thrombi, indicating that other factors, too, must exercise influence. The stream in the femoral vein was often slower than in the saphenous vein, as shown by the fact that there was retrograde filling of the femoral vein in several cases. Frimann-Dahl considers this latter circumstance to indicate that vortices readily occur at the debouchment of the saphenous into the femoral vein, just at the spot where experiences from the autopsy of thrombosis cases have shown thrombi to form.

An author who stresses the importance of the static factor, though for other reasons, is Frykholm (1939). He maintains that thrombosis may occur anywhere at all in the body, but that patho-anatomical investigations have shown four vascular regions to be particularly liable. He cites Rössle and Neumann, and emphasizes that the autopsy results of these authors agree with what he has himself found, namely that the four commonest locations are 1) the plantar veins, 2) the veins of the calf muscles, 3) the veins of the adductor muscles and 4) the veins of the pelvis. He also maintains that, despite the long prevailing view, the femoral vein is very rarely by itself the seat of thrombosis. He considers thrombosis to originate from mechanical injuries to the venous intima, together with slow blood stream. Frykholm considers that, when a patient is lying in bed, there is every reason to suppose that the large venous trunks in the legs are emptied of their blood (cf. varices), so that the lumen becomes no more than a slit. This brings intima into contact with intima, and as the endothelial cells depend in part for their nourishment on the passing blood, they will under such circumstances be exposed to injurious influences. When the blood subsequently fills out the vessels again, some of the cells in the above-mentioned regions will have been injured, and the passing blood will form a small thrombus there. This thrombus gives off coagulating substances

to the blood, which causes further deposits in the direction of the blood stream, since it is constantly receiving substances promoting coagulation. If the stream is swift a white thrombus is formed, since the coagulating substance then have an opportunity to work on thrombocytes and leukocytes on the border. If, on the other hand, the stream is slow, the concentration of these coagulating substances will be strong, and the blood is deposited in layers, one on top of the other, until the vessel obturates. All the blood elements are contained in this latter formation, so that a red thrombus is the result. Frykholm also submits an explanation of the classical thrombus of the femoral vein, with its head, neck and tail parts. He says that when a thrombus is formed in the veins of the adductor muscles, it builds on itself and, following the vena profunda femoris, forces its way into the femoral vein. This part will now be washed over by the femoral blood, which moves rapidly, and in this way a white thrombus will form, gradually blocking the passage. When this has taken place, the blood which collects in the femoral vein distal to the opening of the profunda will try and find its way in a retrograde direction to other anastomoses. The stream then arising is slow, and a red thrombus is formed by means of coagulating substances from the white thrombus. When discussing his material, Frykholm nevertheless points out that though in several of his cases he found a white thrombus in the femoralis vein, there was no red thrombotization of the femoral vein distal to this; he therefore considers there to be little support for retrograde thrombotization in these cases.

Frykholm's theory is well thought out, and seems in many respects probable. As he himself states, however, it has not been possible for him to show that a primary endothelial lesion arises in the way he has assumed. The hypothetical possibility which Frykholm points out must nevertheless be borne in mind.

It is natural to presume that an interference into the human organism, as violent and widespread as an operation, makes for disturbances in the circulation, and it must be a familiar fact that such is the case. Abdominal operations give rise to changed conditions not only for the body generally, but also locally for the viscera. Several authors interpret these conditions as the main cause of that disturbance in the circulation of the blood which arises at operations, and primarily the grave change in it which, in their opinion, gives rise to thrombosis. In the previously cited work by Walters, the author

points out that the diminished circulation in the mesenterial vessels following abdominal operations — at any rate for the first 48 hours afterwards — must be due to deficient peristalsis. Walters considers this condition to make for stagnation in the vena porta, with a similar effect on the entire vascular system. He further maintains that the pain of the wound causes the abdominal respiration to be restricted; this means that the movements of the diaphragm probably become less, and that the normal variations in the intra-abdominal pressure, which are of importance for the circulation, are therewith also reduced. This view is shared by Patey (1929), who maintains that, in X-ray investigations, it is possible to show direct a restriction in the movements of the diaphragm after operations. He considers the decrease in the variations of the intra-abdominal pressure to cause stasis in the veins of the pelvis and the leg. Lockhart-Mummery (1929) takes the same view, which he expresses as follows: 'The cause of postoperative thrombosis is a double one, 1) stasis or partial stasis in the large veins as the result of interference with the movements of the abdominal wall, plus recumbency, and 2) an alteration in the chemical composition of the blood as the result of trauma to the tissues.' He does not support this opinion with any proofs, however.

As has already been mentioned, Frimann-Dahl has made a particular study of the special type of the venous blood stream under different conditions. He has also carried out investigations bearing on the movements of the diaphragm after operations, and shown that they are always restricted after abdominal operations, most markedly on the day after, but then returning fairly rapidly to normal. The retardation of the blood stream in the veins sets in most distinctly from the second day following the operation, continuing during the entire stay in bed. Frimann-Dahl maintains that this circumstance shows that the restriction of diaphragmic movement can scarcely come into consideration as a cause of the retarded venous blood stream. He submits as possible cause of this restriction the meteorism which arises at abdominal operations, and he considers that the restriction of the diaphragm's movements and the meteorism can only set up a transient stasis in the veins immediately following the operation; he attributes the subsequent persistance of the retarded stream to be due partly to the recumbency, with attendant inactivation of the muscular effect, and partly to the reduced amount of blood in the vascular system as a whole.

It may be suitable at this point to touch on the literature treating the *influence of the operation* on thrombosis. As has already been said, the factor constituted by the operation trauma (shock, tissue lesion, anaesthesia) influences *inter alia* the blood stream: relevant literature is therefore examined in this connection, although it does not deal specially with the retardation of the stream, but is intended rather to show the general importance of the operation for thrombosis.

Compilations of fairly extensive materials from clinics of surgery and of internal medicine have been compared for an idea of how the operation influences the occurrence of thrombosis. Many of these compilations do not appear to be very reliable, however, partly because the diagnosis is not clearly formulated—there is confusion of thrombophlebitis, embolism, progressive and local thromboses—and partly because the material has not been treated statistically.

Moreover, the materials for comparison have often been inhomogeneous as regards diagnosis, in that one case presents a clinical diagnosis, another a patho-anatomical one. How great the difference between the real and the diagnosed cases can be is demonstrated by Geissendörfer's work; cf. table 2.

Table 2

Author	No. of fatal lung embolisms	Diagnosed	Not diagnosed	
			No.	% of total
Bodon	58 (med.)	6	52	89.7
Breitner	6	4	2	33.3
Damblé	96	38	58	60.4
Höring	23	5	18	78.3
Kazda, Stöhr	145 surg.	87	58	40.0
Ritzmann after Kirschner	36	6	30	83.3
Total	516	168	348	67.4 ± 2.06

Using compilations of the kind already indicated, several authors have tried to discover whether an operation had contributed to thrombosis. Geissendörfer has sought to clear up this question by the collocation of critically examined statistics, and by calculation with statistical methods. The material used consisted solely of fatal lung embolisms, so that the diagnosis is certain. Geissendörfer denies

any conclusiveness to the figures he obtained, however. Among later authors we may mention Westberg (1937—38) who, after calculations on a material from Upsala, reached the view that the operation trauma unquestionably increases the liability to thrombosis. However, Bauer (1944) shows on a large material collected from a number of different countries that the thrombosis frequency is highest among internal patients, i. e. 2.1 %, and after operation, 1.61 %. This compilation further shows that the number of deaths from pulmonary embolism is also greatest among the internal cases. After partus the thrombosis frequency is 1.01 %, and the frequency of death from pulmonary embolism 3.6 %.

Lively discussion has gone to the varying influence of different operations on the occurrence of thrombosis, and a fair unanimity of opinion has been gained from large materials. Geissendörfer's work (to which those interested are referred) shows that embolisms are rare in strumectomy and hernia operations, while prostatectomy and resection and amputation operations in cancer of the rectum involve a high risk of embolism.

As to the effect of the anaesthesia on the frequency of thromboembolism, the prevalent view, held e. g. by Key, Pirker, Kazda, Killian, Kunz, Sarafoff and others (cit. from Geissendörfer) is that it is small or none.

When discussing the relation of the thrombosis to the operation it is, of course, impossible actually to disregard the importance of age. It is generally agreed that increasing age makes for a higher thrombosis frequency. I do not find it necessary to go into the question more closely here.

Circulatory changes from other causes than those touched on here have been dealt with by Stöhr (1930) and by Bauer (1944). Bauer describes the influence mechanical trauma to the legs exert in upsetting the ordinary circulation in the leg, and shows that thrombosis arises in 12 % of all such injuries, though it is often overlooked. He attributes this to the strictly stationary position of the injured leg and blood-stream difficulties in the vena poplitea on account of haematoma, oedema or tight bandage. Stöhr describes a number of thrombosis cases due to paralysis of the legs owing to encephalomalacia, the cessation of the muscular action of the legs and disturbances in the vascular innervation being advanced as causes. These thrombosis cases are naturally only of theoretical interest in this

connection, but they bear out the view that inactivation of the leg muscles upsets the circulation in the veins of the leg.

Thus, it is obvious that the state of the blood stream must be accorded crucial importance for thrombosis. In general, less attention seems to paid to the type of the stream, more weight being laid on the general circulatory retardation of the blood in the veins of the pelvis and legs, induced by general lesion to the circulation, cardiac disease, recumbency or mechanical stasis. Aschoff nevertheless maintains that the type of the stream, in particular the vortices, is of predominant importance.

Endothelial Lesions

Before Virchow, the main cause of thrombosis was generally taken to be an inflammation. Now, this can readily be assumed to be the case in the septic thromboses, but is less easy to accept as regards the spontaneous ones, which may arise without the slightest manifestation of either local or general infection. It has also been shown by experiment that mechanical injuries to the vascular intima may set up thrombosis at the site of the injuries. Numerous tests have shown that thrombi can easily be induced by lesion of the vascular wall, e. g. cauterization. On the other hand, the investigations described in the previous section have shown that retardation of the blood stream can be assigned a rôle in the formation of thrombi. When the blood stream in a certain vascular region is slow, the metabolism in the cells of the intima may be expected to suffer to such an extent that they begin to die off, whereby a situation comparable to a mechanical lesion is produced, and a thrombus formed. A number of investigators have more or less clearly expressed a view of this kind.

The situation as described in these pages has led to the opposition of two groups of investigators. On the one side are those who consider thrombosis chiefly due to mechanical circumstances (the retardation of the blood stream, and formation of vortices) which cause certain blood elements to be separated off from the main mass and to form thrombi by conglutination or agglutination. The other

side consists of those who assert that the vital factors in the question are those of endothelial lesion and adhesion, the building up of the thrombus being only a secondary phenomenon. These latter investigators consider endothelial lesion to be the crux of the whole problem. The first to reject the Aschoff theory was Ribbert who, as has already been mentioned, gave the lesions of the vascular wall a front place. Regarding the question of adhesion, he says: 'An der Möglichkeit des Haftens hängt aber, ganz allein an ihr, die Frage der Thrombose.'

Klemensiewicz (1917) is an investigator who tried by experiment to confirm the accuracy of Ribbert's view—a view which, as has already been said, was based mainly on theoretical argument and on proofs of a negative kind, i. e. on experiments which argued against Aschoff's theory of vortices. He arranged experiments with cold-blooded animals, primarily salamanders, in which it was possible to see direct under the microscope that thrombocytes adhered to the vascular wall and formed a thrombus when the intima was injured. Klemensiewicz considers that this adhesion cannot be induced solely by lesion of the endothelial cells; a homogeneous fibrin layer must first be formed over the injured cells. He makes a number of experiments to show that a layer of this kind is formed. Thus, he shows how, when blood is allowed to drop on to a well-cleaned slide and is then rinsed off with a liquid inhibiting coagulation (magnesium sulphate solution), a thin, microscopically visible film is left on the slide. He takes this film to be fibrin. The same film can be produced on a glass rod which has been introduced into a blood vessel of a living animal that is to say, from flowing blood in its biologically normal state. Klemensiewicz has also observed fibrin fibres which are attached by one end to the injured vascular walls, and which swim freely in the blood stream and move with it. Plainly, no real film resembling the one on the glass rod was observed here. Klemensiewicz assumed that, when the blood elements adhere to the injured places in a particular way, they are only able to do so by virtue of a net-like film already present on the places in question. There does not, however, seem to be any real proofs of the existence of the film. Klemensiewicz states that, on lesion of the intima, thrombocytes and leukocytes at once adhere firmly to the injured spot. This seems to argue against a previous precipitation of fibrin which would surely take *some* time (cf. ordinary fibrin formation).

For the rest, it may well be questioned whether the observations touched on here would apply to man. Klemensiewicz's experiments are concerned with cold-blooded animals, which may be supposed to react differently from warm-blooded ones. Apart from this, there are no sure proofs that a thin fibrin layer is first formed, and that the corpuscular elements of the blood then adhere to it, gradually forming a thrombus. All that the experiments described actually show is that thrombi arise on lesion of the intima, and this was a long-familiar fact already.

Dietrich (1920) also claimed to have proved lesion of the intima to be the main cause of thrombosis by the investigations he made into the connection between thrombosis and war injuries. He states emphatically that, in all cases of thrombosis, lesion of the endothelial cells from infection is the fundamental cause. In proof of this view he quotes a large material of thrombosis cases among the wounded during the Great War 1914—18. He shows that it is only exceptionally that thrombosis occurs in the ligated vascular stumps after amputation, provided there is no infection present at the same time. He furthermore submits examples of thromboses which, with no visible connection with the infected wound, appear on the favourite site of the spontaneous thromboses; on the strength of this he draws the conclusion that the former are to be regarded as true progressive thrombi. According to Dietrich, a careful investigation of these cases always shows that infection can be demonstrated on the site of origin of the thrombosis, or in the actual thrombus. In other cases he considers a change of the composition of the blood to be present, and that thrombi are formed as a result, though only at places where there are suitable lesions, caused by local infectious changes. On the reasons given here, Dietrich has reached the view that infection is always the main cause of thrombi, and he makes the following summarizing pronouncement: 'Die Thrombose stellt sich demnach, unter Heranziehung bereits früher gewonnener Anschauungen nicht nur als ein mechanischer, sondern als ein reaktiver Vorgang dar, der ausgelöst wird durch eine Störung im Verhältnis von Blut und Gefässwand, sei es dass die Schädigung der Wand oder die Veränderung des Blutes stärker hervortreten lässt oder im Zusammenwirken beider keine Entscheidung ermöglicht. Eine Behinderung der Blutströmung tritt nur als Begünstigung und formgestaltende Bedingung hinzu.'

A possible objection to this view is that the thrombi under discussion here are not spontaneous in the actual sense of the word, so that they do not warrant conclusions of the kind Dietrich drew from them. The thrombi he describes set in among severely infected subjects. (One case even showed both typhus abdominalis and dysentery, as well as the infected wound.) It can hardly be justifiable to compare a spontaneous thrombosis in a case like this with one in a pure case, e. g. a patient operated upon for inguinal hernia.

Another to subscribe to the theory of endothelial alteration from different causes as the main cause of thrombosis is Ritter (1926). Nevertheless, his view diverges from that of Dietrich in so far as Ritter takes the disturbed metabolism (inanition) of the endothelial cells to be the primary factor in the problem. In a large work this author describes the experiments he submits to prove the accuracy of his theory. He uses a special endothelial reaction, worked out by Gräff (cit. from Ritter), that shows even negligible changes in the normal cell picture. By injecting different unspecific protein substances into the vascular system of the experiment animal, he is able to demonstrate that his method sets up a difference in the appearance of the cell, whereas no change can be observed in the customary histological investigation. The injection of a mercury chloride solution gives practically at once a very strong cellular change but no coagulation of the blood, though this last might be expected after so powerful a poison. An investigation of the animal's vessels after 6 hours showed incipient thrombosis, on the other hand. From these experiments (complemented with a number of others, of no interest in this connection) Ritter draws the conclusion both that unspecific protein substances can change the metabolism of the endothelial cells, and that the first beginnings of the thrombosis are to be sought in the cells of the intima. He maintains that the protein substances used at the experiments can be compared with those substances of similar nature which, in man, are introduced into the blood from large haematomas and pus cavities.

In a new work (1929) Dietrich advances several other of his experiments as proofs of the theory he had laid down earlier. Thus, he sensitized rabbits by intravenous injections of bacterial toxins and bacterial cultures repeated at varying intervals. A microscopic examination of the intima of such animals showed it to have ab-

sorbed the bacteria, with great increase of cells; at the same time a thin homogeneous layer separated off, which Dietrich considers to be identical with Klemensiewicz's fibrin film. Dietrich considers this membrane to arise from the joint action of the endothelium and the plasma flowing past it. He further maintains that both the endothelial cellular increase and the thin fibrin film seen in the experiment animals can also be observed in human subjects suffering from diseases associated with thrombosis.

The theories of Dietrich and Ritter that endothelial changes are the primary factors in thrombosis have been violently attacked by many authors. These latter do not deny the existence of changes of the kind Dietrich and Ritter claim to have shown, but they question whether the experiments on which this view is based can be applied indiscriminately to man. Aschoff (1934) points out that these experiments can in no way be equated to the conditions prevailing at the onset of spontaneous aseptic thromboses, to which, therefore, the conclusions drawn cannot justifiably be applied. Aschoff further points out that the cellular increase in the intima which Dietrich points out, and which may sometimes occur in man, has nothing to do with sensitization. Cellular agglomerations of these kinds are purely an age phenomenon, as Lucadou has shown (1933). Geissendörfer (1935) will not even allow the endothelial reaction to be of importance for thrombosis. To explain this view he submits an investigation based on the findings of different investigators. Thus, he points out that many diseases, such as scarlatina, sepsis and other infectious conditions, show the same endothelial changes as those indicated above, without any particular frequency of complicating thrombo-embolism. He also says that, in many diseases, large quantities of protein are introduced into the blood channels for therapeutic ends, e. g. serum injections in diphtheria and tetanus, without a very great frequency of thrombosis among these patients subsequently. Another circumstance which Geissendörfer cannot reconcile with Dietrich's view is that the progressive vein thromboses are especially easy to detach from the vascular wall, which hardly suggests any very intimate connection between thrombus and wall.

Over and above what the authors mentioned here have adduced against Dietrich's and Ritter's view of the primary importance of the endothelial cell for the genesis of thrombi, we may also men-

tion a work of Klotz (1932) who has shown that all operations injure the endothelial system to a greater or lesser extent.

Tannenberg (1937) also rejects the view that endothelial change is the main cause of thrombosis. This investigator has induced thrombi by means of different experiments, but he has never been able to find the fibrin film described by Dietrich and, earlier, by Klemensiewicz. Concerning Klemensiewicz's membranes that are formed if the blood dropped on to a glass slide is rinsed off with certain salt solutions, Tannenberg showed that the same membrane can be obtained if serum is used, so that there is obviously no question of fibrin.

A work by Takaura (1937), describing certain experiments promoting thrombosis, shows that different organ extracts induce a certain swelling in the endothelial cells of the vessel on injection. Takaura also shows that when a number of experiment animals are injected with fresh or boiled organ extract or histamin, they all show retardation of the blood stream and lesion of the endothelial cells in the intima, but that thrombosis occurs only when the fresh organ extract is used. This shows that it cannot be the change in the vascular wall which is the main cause of thrombosis.

Another investigator, Kaneko, tries (1939—1940) to illustrate by experiment the part played by infection in thrombosis. He uses bacteria cultures, brushed on to an exposed vein to induce thrombosis. Kaneko investigates the changes in the blood which occur simultaneously with the formation of the thrombus. He found in so doing that, when thrombi are induced in this manner, the composition of the blood is very little changed. The case is different if thrombi are induced by means of injections of different bacterial toxins by the intravenous route. The staphylococcus toxin, for example, can set up thrombosis, while the streptococcus toxin cannot. In both cases, however, changes in both endothelium and blood pressure are set up and, when the staphylococcus toxin is used, an increased tendency in the blood to coagulate, as well. If a substance increasing this tendency is first injected, thrombi can be induced with streptococcus toxin also. Only in exceptional cases do thrombi result from the coagulatory substance alone. From these experiments, Kaneko concludes that injury to the endothelium does not greatly influence the formation of thrombi.

It appears from the investigations reviewed here that thrombosis can be induced by the experimental infliction of injuries to the endo-

thelium. It is possible that such injuries are also present when thrombi occur 'spontaneously' in man. This has not been definitively proved, however. It has been shown in the earlier section that a retarded blood stream and mechanical conditions exert material influence, and it is possible that – at least in a number of cases – an endothelial lesion helps. Furthermore, it is obvious that as a thrombus blocks the circulation through a vessel it should, secondarily, produce a nutritional derangement which is not easy to differentiate from a possible primary lesion in the endothelium.

Blood Changes

Before the emergence of modern surgery the discussion of the thrombosis problem was on the whole centred round the two factors: retardation of the blood stream and lesion in the vascular wall. The third factor, blood changes, receded rather into the background. This was certainly not because it was considered less important, but rather because suitable methods and technical possibilities of investigating the different constituents of the blood were not available at that time.

With the progression of surgery, one of the complications following operation, i. e. thrombo-embolism, came to attract more interest than formerly, and to stimulate investigation into problems relative to it. In due course methods were also worked out, making it possible to determine blood changes in a simpler way and more rapidly than before. This meant that interest was led into these channels.

An important point in this connection is the time after the operation for the occurrence of the thrombosis. The spontaneous thrombus is a process which must of necessity take some time to form. To begin with, of course, the thrombus yields no clinical symptoms; these only appear when the vascular lumen has become occluded, or if an embolism of a certain magnitude has arisen before this. A certain amount of light should be obtained from seeing how long after the operation the occlusion and the embolism respectively become clinically manifest. In view of what has previously been said about the difficulty of diagnosis, the lung embolism cases in particular should be confirmed by autopsy. Large thromboses localized in the legs can gener-

ally be diagnosed clinically. There is nevertheless a possibility that a number of thromboses are certainly never diagnosed, and therefore are not included in the material, but this circumstance is of no consequence here.

Table 3, compiled from the reports of several authors, illustrates how the risk of embolism stands in the time immediately following the operation:

Table 3

Author	No. of embolisms	Time of embolism after operation
Hermansson	50	between 1— 7 days 6 emb. (after partus 0 emb.)
		» 8—14 » 20 » (» » 7 »)
		» 15—28 » 15 » (» » 2 »)
Nicolaysen	210	Mostly within 10 days. Cannot be calculated, as figures not given. Diagrammatic curve.
Scheidegger	143	between 1— 5 days 31 embolisms
		» 6—10 » 25 »
		» 11—15 » 24 »
		» 16—85 » 63 »
Westberg	64	» 1— 5 » 15 »
		» 6—10 » 29 »
		» 11—20 » 20 »
Linde	230	On an average on the 10th day.

If a calculation of these figures is made, it will be seen that, of 257 embolisms, 106 occur within 10 days after the operation, i. e. almost half. If Nicolaysen's and Linde's cases are then added, this figure becomes even larger. And, indeed, thromboses are usually diagnosed at this time. Mention might here be made of a compilation of the present author's (1940) of a 10-year material, in which it is shown that, of 91 clinical thromboses, 74 manifested themselves between the 6th and the 11th day. These figures make it difficult to determine when thromboses arise. It may be assumed that the embolism is an incipient thrombosis, which is torn from the vascular wall before it has been built up into a manifest thrombosis. In other cases it consists of detached portions of an already obturating thrombosis.

As embolisms are of so varied occurrence, but are nevertheless most frequently found within the first 14 days after the operation, these facts rather suggest that there is a causal connection between the thrombosis—embolism and the operation trauma.

The factors discussed in the previous pages, i. e. reduced rate of stream and lesion of the vascular wall, have already been shown to influence the occurrence of thrombosis, but they were not able fully to explain it, and there was only sketchy knowledge as to how far further factors are involved. The blood changes therefore came more and more to the fore as objects of study, particularly those occurring after operations and childbirth, and attempts were made to link them up with the postoperative and puerperal thrombosis. A large number of investigators have gone into these matters. Some have directed their attention solely to variations in morphological respects, others have demonstrated changes in the suspension stability, still others have investigated changes in the content of certain blood constituents.

The following pages will discuss the most important available literature on these questions; this will, however, call for a systematic division. The discussion has dealt primarily with morphological blood changes, then with changes of the plasma composition (the albumin-globulin ratio) and the changing content in the plasma of the substances reacting on coagulation (thrombokinase, prothrombin, thrombin and fibrinogen, as also antithrombin), and finally with the variation in the blood's coagulation time.

Investigations bearing on the relation between blood changes and thrombosis are, quite naturally, scarce, while the works dealing with the postoperative and puerperal blood changes are many. As they throw light on the condition of the blood during the time that thromboses customarily set in, it is important that they should be discussed in this connection.

Morphological blood changes, i. e. variations in the number of the corpuscular elements, as well as in their way of reacting, occur in different diseased states. As the separate form elements in the blood participate in thrombosis in varying numbers, and as they also vary in number in the different sections of the spontaneous thrombus, it is natural that attempts have been made to place these variations in relation to the occurrence of thrombosis.

The *erythrocytes* seem to be of little, if any, importance in thrombosis. It has already been shown that there are absolutely no red blood corpuscles in the head part of the thrombus, whereas there are a number interspersed in the neck part, and they occur in the tail part in the same proportion as in the flowing blood. These circumstances make it hardly likely that the erythrocytes should play any part in the occurrence of thrombosis. Kristenson (1928) has shown that the erythrocytes do not show any noteworthy changes in number in association with thrombosis.

As regards the *leukocytes* it is necessary to note not only the number and the relative proportions in the different types (the differential count), but also their adhesive tendency. The first to study the changes in the leukocyte count following operations was Schulz (1893), who induced a post-operative leukocytosis in animals by aseptic measures. This investigator considered that the cause of the increase in white blood corpuscles was to be sought in 'the irritation' from the wound, and not in an infection. To explain this he adduced that, when an infection had been set up in experiment animals by injecting bacteria into their vessels, an initial decrease of the leukocytes with subsequent leukocytosis could be observed. There was no decrease after aseptic operations. Several investigators have subsequently verified these findings on man. Stahl (1929), for example, has shown that the same phenomenon arises after operations upon human material, and he considers resorption of protein substances from the wound to lie behind the leukocytosis. This author also found immature forms among the leukocytes. Löhr (1923) investigated the number of white blood corpuscles before and after operation in 37 cases, the differential count being also made in 29 of them. He found in so doing that all cases showed a more or less pronounced leukocytosis, with its highest value after 6—24 hours. He was, in addition, able to establish that all the investigated cases presented immature forms of leukocytes ('Linksverschiebung' of the picture according to Arneth—Schilling). Allen (1927) also studied the varying number of the leukocytes after operations, and his diagrammatically presented values confirm Löhr's findings. Others to study these matters are Eufinger and Kiltz (1935); they showed that leukocytosis sets in 3 hours after emergence from a narcosis. There were normal values as early as after

24 hours, but if there had been an operation the leukocytosis persisted longer, and normal values could not be found until 4—5 days after the operation. The primary value in 15 narcotized but not operated cases was, after calculation, mean = 8 200 ± 600; 3 hours after the end of the narcosis the value was mean = 15 000 ± 400, so that the difference is also statistically significant.

Thus, the investigations made into the number of leukocytes as affected by operations show that leukocytosis is set up both by the surgical intervention and by the narcosis. As the leukocyte count is also influenced by such factors as the partaking of food, infection etc., it is not likely that any regular relation to thrombosis exists—nor has it been possible to find any such.

As early as 1892 Hankin observed a previously unknown property in the white blood corpuscles; namely, their power of adhering to firm objects. Hankin points out that as soon as the blood leaves the vascular system, the leukocytes become highly viscous. A proof of this is Hankin's observation that a glass rod, introduced into a glass beaker containing fresh hiridunized rabbit blood, was invested with adhering white blood corpuscles. This observations was afterwards verified by several later authors. Thus, Ponder (1908) builds on it when working out a method for the study of living leukocytes. In studies bearing on the adhesive tendency of the leukocytes, Wright (1921) and Bond (1924)—quoted in both cases from v. Philipsborn—claim to have found that more leukocytes adhere during the fever-free stage in sufferers from malaria and lymphogranuloma than in periods of fever.

The first to work out a method for determining the variation in the adhesive tendency of the leukocytes, and to investigate these matters in any detail, was v. Philipsborn (1930). This method, which will be discussed more closely in a later section, can be briefly described as follows. A blood drop of a certain size is placed on a ground glass slide, of non-alkaline glass, and is left to coagulate in a moist chamber and remain for half an hour at 37° C. The drop is then rinsed off with a jet of water, and the slide is left to become air-dry. By counting the number of leukocytes adhering to the slide in 100 small squares in a net micrometer, dividing the sum by 10 and taking the ratio of the mean of 2 calculations to 1000 white blood corpuscles in the blood, a value is obtained which v. Philipsborn calls 'Klebrigkeitswert'. He determined the value in 6 healthy men and

made 10 determinations, obtaining a lowest value of 5.5 and a highest value of 8.2. Determinations from the same man on different days show no values outside the limits given here. Furthermore, v. Philipsborn has calculated the adhesion value in diseased men, and in both healthy and diseased women. The number of cases is small, as is also the number of determinations made, so that the reliability of the method can hardly be assessed here. A calculation of 10 values given yields a mean of 6.5 for men, but it should be mentioned that half of these values refer to the same person, whose mean is 6.8.

Lampert (1931) is opposed to v. Philipsborn's view of the leukocytes' adhesion tendency. He attributes this tendency solely to varying forces of surface tension, which vary with the material used. He showed (with photographs) that there is a great difference in the number of adhering leukocytes if he applied v. Philipsborn's method to objects of glass (with great 'Benetzbarkeit', by which he implies the tendency of fluid substances, and above all of water, to adhere to firm surfaces) and of amber (with small 'Benetzbarkeit'). He criticizes the proof v. Philipsborn gives for his statement that chemotactic forces cannot be in question, namely, that the adhesion value of glass and quartz is the same. Lampert points out that this is no proof, since quartz and glass have almost the same 'Benetzbarkeit' for water. These objections are only objections to the theoretical basis of the method, however; the fundamental idea that different bloods on one and the same slide give divergent values remains unshaken. No explanation of this changing adhesion tendency of the leukocytes has as yet been forthcoming. Attempts have been made to place the adhesion value in relation to other blood changes and thus obtain a common basic cause, but no convincing solution to the problem has been reached.

In a work of 1938 Bayer and v. Philipsborn answer Lampert's criticism; they maintain that the resistance the latter took to be physical does not argue against the findings v. Philipsborn advanced, if it is remembered 'dass die Leukocyten ihre eigentliche Aufgabe erfüllen, indem sie zu Grunde gehen', and if, furthermore, it is assumed that irritated leukocytes whose functionary powers are enhanced are more easily injured and die off, and are therefore more readily affected by the surface forces of the slide. Bayer and v. Philipsborn also consider that the adhesion tendency cannot be due

to chemotactic forces, since objects of quartz and glass give values which lie within the method's limits of error. This statement does not seem to be proved by the experiments they performed for, according to Lampert, quartz and glass afford water the same opportunities of adhesion. However, neither Bayer and v. Philipsborn nor Lampert say whether the quartz is crystalline or amorphous, a detail which might be thought of importance to the water adhesion. In the above-mentioned work, Bayer and v. Philipsborn communicate that the standard error of the method (calculated on 30 determinations) is $\pm$ 0.4, i.e. somewhat higher than the one reached by calculations on the figures from other authors (cf. Eufinger and Kiltz in the following pages). Bayer and v. Philipsborn claim to have found that the power of adhesion shows rhythmic changes throughout the 24-hour cycle, with the maximum about 6 p. m. and the minimum about 3 a.m. As the number of investigated persons is not given, and as no figures are submitted beyond a graphic curve, there is no way of judging this statement. The same is true of investigations as to the changes of the adhesion value in connection with exertion, varying air pressure and climatic variations.

The question of the connection between the adhesion value and other blood changes has been dealt with by several authors. Scholz (1933), for example, studied the adhesion tendency of the leukocytes and at the same time also determined the sedimentation rate in a material consisting of pregnant women. He says that no connection can be shown in his material between these two, since the adhesion value is subject to variations to which the sedimentation rate is not. The latter rises to a certain value in the 4th month, and remains there until the end of the puerperium, whereas the former reaches its maximum with partus. Kanai (quoted from Vejlens) has studied the adhesion between leukocytes and carbon particles in phagocytosis experiments; he found it to be increased in a medium rich in fibrinogen and also, though to a lesser degree, on a rise in the serum globulin. Vejlens (1938) has carried out investigations *in vitro* on human blood and studied the effect of an addition of gelatin (1 %) on the adhesion powers of the leukocytes; it was nil. On the other hand, Vejlens was able to show that the adhesion is inhibited by the addition of hypertonic sodium chloride solution. He considers the adhesion to be due to the amoeboid movement of the leukocytes and their power of attaching themselves to firm surfaces. According

to Vejlens, it is unthinkable that so many leukocytes could collect at the glass surface during the short time required for the coagulation of the blood, unless they emigrated there. He leaves this question open, however, though he points out that, when the amoeboid movement is inhibited (addition of hypertonic sodium chloride solution does this, according to Thoma), the adhesion is also inhibited. Vejlens has also made the experiment of rinsing slides bearing attached leukocytes with 1—2 % NaCl solution; he found in so doing that practically all the leukocytes gradually came loose and disappeared. He links this up with the observation that hypertonic sodium chloride solution counteracts the effect that fibrinogen and other hydrophile colloids have on erythrocytic aggregation, decreasing the rouleaux-formation. Vejlens has also studied the form of the different attached leukocytes at varying contents of fibrinogen in the blood. He demonstrates that blood with low suspension stability (from pneumonia cases) or normal blood to which fibrinogen or gelatin has been added, yield preparations where a large number of the attached leukocytes are provided with long thread-like offshoots. Vejlens considers that this type of cell originates from one part fastening to the glass, the other to the clot, and when the latter retracts, the cell is pulled out like a piece of chewing-gum and forms these offshoots. He also thinks that this experiment suggests that a lowered suspension stability (fibrinogen and gelatin added to the blood) increased the adhesion tendency.

In Scholz's above-mentioned work the sedimentation rate, which is a measure of the stability in the erythrocyte suspension, is shown not to change on those occasions when the adhesion value rises considerably (verified statistically).

Eufinger (1932) has used v. Philipsborn's method to investigate the adhesion values in women; he claims to have found that, during the intermenstruum, there are normal values (the same as in men), but that immediately before menstruation there is a marked decrease, rising during the actual bleeding to fall again a short time after the end of menstruation to subnormal. Eufinger investigated 22 healthy women and reports that all showed changes in connection with menstruation, of the kind described here. He gives no primary figures, so that the standard error cannot be calculated. The adhesion value has also been determined in pregnant women. The primary figures in these investigations are to be found in a work by

Scholz (1933), and they can therefore be used for calculation. Ten determinations made on healthy women gave the mean of 5.7 ± 0.5, and a comparison with 29 cases determined immediately before partus yielded a mean of 12.1 ± 0.4. The difference, 6.4 ± 0.61, is consequently statistically significant. The same is also true of the figures for the labour stage, when the adhesion value is further raised. At the same time as he determined the adhesion value, Scholz carried out determinations of the leukocyte count, the differential picture and the sedimentation rate. A survey of the figures in the table shows that the sedimentation rate runs fairly parallel with the adhesion value in that the former can be seen to rise up to the 4th month, but that after partus a high sedimentation rate remains, while the adhesion value falls to normal. Nor can any connection be found between the leukocyte count and, on the one hand, the blood picture and, on the other, the adhesion value. There are normal leukocyte counts, for example, in the 10th month, while the adhesion value, as already indicated, shows a rise compared with the normal. Scholz stated that the coagulation time of the blood is connected with the adhesion value inasmuch as high adhesion values are combined with low coagulation time, and *vice versa*. As no figures are given, however, the full weight of this statement cannot be assessed.

Eufinger and Kiltz (1935) have made a close study of the adhesion value as affected by narcosis and operation. As has already been mentioned, they investigated the leukocyte count and adhesion value before and after narcosis, and showed that the former changed in a typical way, rising 3—4 hours after the narcosis and returning to the original value after 24 hours. The adhesion value, on the other hand, did not change at all. This statement can to a certain extent be verified statistically. The calculation of the adhesion value before the narcosis (15 cases) gives a mean of 5.8 ± 0.3, and one of 5.5 ± 0.3 is obtained 3 hours after the end of the narcosis; the difference between the two series is 0.3 ± 0.4, implying no significant difference. The authors conclude from these investigations that the number of leukocytes bears no relation to their adhesion tendency. Eufinger and Kiltz have also made determinations of the adhesion value before and after operations; they report that it sinks to a minimum about 3 hours after operation, and then rises, with its maximum on the 3rd—4th day. The investigation comprised 17 cases of aseptic operations; a statistical calculation of the submitted

figures shows, before the operation, a mean of 5.3 ± 0.3, and one of 4.8 ± 0.2 at the time for the minimum, indicating that the divergence of 0.5 ± 0.3 is not significant. Figures from the material quoted here do not, therefore, warrant the conclusion that the authors advanced. The statement that there is a maximum on the 3rd—4th day seems to be justified, since the figures for the 3rd day yield a mean of 9.6 ± 0.8, making the divergence 4.3 ± 0.8. The material is small and the difference between the different series is not large, so that the figures quoted here should, despite the statistical verification, be appraised with caution until larger materials are submitted.

Recent works by Swedish authors (Holmgren and Wilander, 1937, Jorpes, Holmgren and Wilander, 1937, Holmgren, 1938) concerning the tissue mast cells producing heparin have drawn attention to the *basophile cells* of the blood.

Holmgren (1940) shows that human eyes enucleated for thrombosis in the afferent vessel present a striking decrease of the mast cells. Holmgren considers that a similar state of affairs may attend thromboses in other vascular sections. However, as he himself points out, this may equally well be cause as effect. These investigations refer to tissue mast cells, but cells of the same type are also encountered in the blood, though they are few under normal conditions. Their real nature is as yet unknown. The granules present in the blood mast cells have the same capacity of staining with toluidine blue as those in the tissue mast cells, suggesting that these granules consist of heparin, or some similar substance (Hedenius 1940). A circumstance worthy of notice in this connection is that the coagulation time of blood can be prolonged in cases of myeloid leukaemia (Holmgren and Wohlfart, 1941) where there is an increase in the blood mast cells.

In the earlier experiments into the mechanism of thrombosis, Bizzozero (1882) showed that the thrombus induced by a needle puncture in a mesenterial artery of rabbit consisted exclusively of *thrombocytes*. Eberth and Schimmelbusch (1888) carried out experiments with the same end in view, but they induced thrombi in coarser vessels. Using a thread, they heavily ligated the femoral artery of a dog and left it for quarter of an hour (intima lesion); they then loosened the ligature and let the blood flow free in the vessel for three quarters of an hour. This gave them a good thrombosis, mainly

formed of thrombocytes. These experiments have subsequently been verified many times by different investigators.

Eberth and Schimmelbusch also studied the blood stream in respect of the localization of the form elements in it. A direct study under the microscope of the course of the blood through the vessels of living animals (omentum vessels) showed them that the different corpuscles followed certain laws in relation to one another and to the vascular wall. The middle of the blood stream is taken up by a broad red current, and at the sides is a thinner zone, consisting of plasma. This state of affairs is particularly pronounced in the veins and the capillaries. The plasma zone is broadest in veins, and in capillaries with the exception of the very narrowest ones, where the lumen only allows the passage of single blood corpuscles; in these capillaries no partition between axial stream and border can be observed, nor any plasma zone. When the blood flows at a normal rate, no details in the axial stream can be distinguished, but white corpuscles can be observed here and there in the plasma zone; these last appear as it were to roll slowly along the vascular wall. The authors mentioned here take this picture of the blood stream to have a purely mechanical cause. They attribute the distribution of the corpuscles in the blood stream to variations in the specific gravity of the elements, opining that the heavier ones keep in the axial stream and the lighter ones keep along the borders, where the stream is slow. If the rate of the blood stream is reduced, one gets considerably more leukocytes in the border zone, and if it is retarded still more, thrombocytes range themselves along the edges of the vessel at the expense, to a certain extent, of the leukocytes. Total stagnation of the blood is accompanied by an ununiform distribution of the blood elements. On the formation of vortices in the current, however, leukocytes and thrombocytes are forced out into them in large numbers.

This view of the intravasal course of the blood stream was the accepted one until Fåhraeus (1928) showed, by a number of different experiments, that both the theory as to the disposition of the different blood corpuscles in the blood stream and the opinion as to the active cause (the specific gravity) were wrong. He verified that the blood stream showed an axial, rapidly flowing part and a more slowly flowing border part, practically stationary in the zone nearest the vascular wall. Both the white and the red corpuscles are transported in the axial stream, the white fastest, on account of their size. Fåhræus

showed that it is the varying size which decides where the blood corpuscles are placed in the stream. When it is slow, or when the suspension stability is upset (globulin increase), the erythrocytes tend to form large aggregates, which then, on account of their superior size, force the leukocytes into the border zone. Fåhræus makes no special statement about the thrombocytes, but as they are small in relation to erythrocytes and leukocytes, they should under normal conditions flow at a great distance from the axial part of the stream and, when the stream is slow or the suspension stability upset, be pushed still further out towards the part of the border zone close to the vascular wall.

As has already been mentioned, Aschoff's works on the histological structure of spontaneous thrombi in man have made it clear that the white thrombus consists mainly of thrombocytes.

Since, then, the thrombocytes are the blood elements constituting the first foundations of the thrombi, forming in addition the bulk of the head part, they have been intensively taken up by present-day investigators, also, in connection with the thrombosis question. It is, of course, reasonable to assume that their varying number or variations in their manner of reacting might throw light on the way the organism is predisposed to thrombosis, or provide proof that thrombosis had developed (occult thrombosis).

Hueck (1925) was the first to make calculations of the thrombocyte count in surgical cases. He investigated 100 different cases of this kind, but could not find any variations, as these lay within the limit values of the method.

Normann (1928) investigated the thrombocyte count before and after operation; his material (60 cases) shows that a thrombocytosis always sets in, usually between the 10th and 16th days, its intensity depending to a certain extent on the magnitude of the operation: an operation for hernia gives a less pronounced increase of thrombocytes than a stomach resection, for example.

Using a fairly large material, Windfeld (1933) carried out investigations into the changes of the thrombocyte count in surgical diseases. He used the method Petri worked out on rabbits, and set its standard error at 6.3 % by means of determinations on healthy human subjects. He investigated the thrombocyte count in healthy subjects, and found twenty-four-hour, monthly and yearly variations, which nevertheless all lie within 3 times the standard error. Wind-

feld gives his figures from investigations into operation cases (67 cases) by graphs, so that an approximate calculation only is possible, but it nonetheless shows there to be significant differences before and after the operation, i. e. values varying from the pre-operative ones by more than 3 times the standard error. He shows that a post-operative thrombocytosis begins on the 4th—5th day, with a maximum on the 10th—15th day. After this time the values slowly fall to normal. Windfeld found that the narcosis does not influence the thrombocyte count. Comparisons between different operations, such as resection of the stomach and of the thyroid gland, and in fracture cases, show the curves for the thrombocyte count to follow courses resembling one another. Windfeld has also endeavoured to find a relation between loss of blood and variations in the number of thrombocytes. However, he has found that, in several cases (the number is not given), it is possible to see that the thrombocyte count does not rise after considerable blood losses, while it may increase after quite slight ones. This has also been established by Petri (quoted from Winfeld) in investigations on rabbits. Even extensive phlebotomy did not occasion a rise in the thrombocyte count. Windfeld took these circumstances to suggest that the rise of the thrombocyte count cannot be explained by a regeneration of the thrombocytes which have developed beyond the normal value. A circumstance in favour of this view is that fracture cases show the same rise as operation cases. On the strength of the argument reviewed here, Windfeld considers the increase of thrombocytes after operation to be due to a resorption of products from cellular degeneration.

Sulgar and Boszin (1929) have also interested themselves in this problem. They report from 30 cases that, after operation, there was an increase in 14, a decrease in 6 and no difference in 10. Evans (1929) communicates that there is an increase of the thrombocytes on the 8th—16th day after an operation. He gives no figures in his works, so that their data provide no sure guidance.

The reports as to the thrombocyte count after operation are consequently somewhat divergent. Normann finds an increase on the 8th—16th day, Windfeld on the 10—15th day and Evans, finally, on the 8th—16th day. Sulger and Boszin, on the other hand, could not find any significant increase. The only two who give calculable figures based on an adequate material are Normann and Windfeld. Their works show a thrombocytosis to set in after operations.

Studies have been made of variations in the thrombocyte count during the puerperium, also. As early as 1907 Rebaudi states that there are changes in the number of thrombocytes during pregnancy, and that there is a postpartum increase with a maximum 4—5 days after delivery. Schilsky (1921) submits smaller variations during this period, while Horwitz (1927) could not show any change in the number of thrombocytes at all. This latter author used Hoffmann—Flössner's method of counting which, according to Hartmann (1932), is uncertain. Thus, pronounced falls of the thrombocyte count in haemorrhagic diatheses cannot be demonstrated with this method, though they can with that of Kristenson. Schulte (1928), Evans (1929), Hare and Glynn (1929) and Krukenberg (1930), maintain that thrombocytosis appears after partus. The time for the maximum value varies, however; Schulte finds it to occur on the 5th—6th day, Krukenberg not until the 9th—12th day. Using Fonio's method, Radvány (1929) has calculated the thrombocyte count in 101 women during pregnancy and partus, but cannot find any divergencies. The authors mentioned here have illustrated their findings by graphic curves, or else advanced their results without this support. This means that the statements submitted cannot be assessed. The varying results may be due to the use of different methods in the different investigations.

Kristenson (1938), who worked out the method of determining the thrombocyte count now used in Sweden, investigated a large material at the gynaecological clinic in Upsala. He showed statistically that there is little variation in the number of thrombocytes during the first days after partus, but that there is a subsequent increase with its maximum on the 5th—7th day.

Different experiments have made it clear that the first rudiments of the thrombus consist of a thrombocyte clumping. Furthermore, as has already been indicated, the histological picture shows the thrombus to be comprised mainly of thrombocytes. It is thus obvious that the question of the genesis of the thrombus is intimately linked up with the thrombocytes, above all with their tendency to agglutinate. It is therefore natural that this property in the thrombocytes, which seems to be the nucleus of the whole problem, has come to interest a number of authors and become the object of detailed study. Attempts have also been made to connect an increased agglutination tendency with thrombosis.

Zurhelle (1909) studied experimentally induced thrombi formed by agglutination of the thrombocytes. He makes no pronouncement as to changes in this tendency, only claiming to have found that these conglomerations have no fibrin and are therefore products of a process separate from the fibrinogen coagulation.

Starlinger and Sametnik (1927) studied the agglutination tendency of the thrombocytes under varying conditions. Thus, they placed blood (120 different samples) in an electric field and studied the behaviour of the thrombocytes. They found that all thrombocytes migrated to the anode, concluding from this that they must be negatively charged. The iso-electric point for thrombocytes was found to be lower than that of the globulins but higher than that of the albumins. In blood with a high globulin content the thrombocytes migrated towards the anode more slowly than when the globulin content was normal that is, their electrostatic charge decreased and the tendency to agglutinate was thereby enhanced. These *in vitro* experiments show that displacement of the plasma colloids towards the globulin side *in vivo* set up favourable circumstances for the agglutination of the thrombocytes.

Barta and Jakab (1929) have also studied the agglutination of the thrombocytes experimentally. To pure isotonic thrombocyte suspension they added albumin, globulin and fibrinogen solutions of the same concentration in the ratio of 10: 2, and found that the fibrinogen solution produced the strongest agglutination. By adding thrombocyte suspension to solutions of different p_H Barta and Jakab were able to establish that agglutination also took place in a protein-free medium and at a p_H between 3.8 and 4.3. As, however, the p_H is higher *in vivo*, a study was also made of the agglutination tendency in different protein solutions with varying p_H. It was then found that the agglutination was strongest at a p_H where the protein flocculation was greatest. Thus, agglutination occurs in the presence of protein substances, above all fibrinogen, both at a low and a high p_H. The tendency for the thrombocytes to agglutinate increased with the fibrinogen content. Furthermore, Barta and Jakab maintain that the proteins adsorbed on the surface of the thrombocytes impart to them the same properties of precipitation which they themselves possess. The communicated experiments admittedly build on a subjective method of appraising the agglutination, but

the differences between the values in the different experiments are so great that they should prove the point.

Lenggenhager (1934—35) opposes the hypothesis advanced by several authors that the agglutination powers of the thrombocytes differ as to degree. He assumes that the thrombocytes have no 'Klebrigkeit'. He points out that thrombocytes can remain in suspension in citrated plasma for several days. Lenggenhager attributes the aggregation of the thrombocytes to their affinity for freshly formed fibrin. He supports his view with several of his own experiments, which, however, in the light of later works do not seem to deserve notice and are therefore not reviewed.

It may be mentioned that Lenggenhager has succeeded in producing agglutination of kaolin granules, suspended in thrombocyte-free citrated plasma, by the addition of serum. When a citrated plasma-kaolin suspension of this kind has been totally defibrinated by successive addition of serum (thrombin), the kaolin granules become coated with a thin layer of a substance which Lenggenhager takes to be fibrin. He considers this opinion of his is supported by the fact that the remaining solution cannot be made to coagulate; all the fibrin is on the kaolin granules. He derives further support from the circumstance that the granules possessing this layer are heavier, with greater tingibility and powers of charring. Lenggenhager therefore asserts that the fibrin formed in the flowing blood has a special affinity for the thrombocytes. After all, however, his own experiments have shown that fibrin has as much affinity for kaolin granules as for thrombocytes.

Nevertheless, Fonio and co-workers (1942) have been able to make *in vitro* studies of the thrombocytes and their importance for the coagulation under the microscope direct (darkfield illumination). These authors are able to show on excellent microphographs both that the thrombocytes appear rounded in states of rest and with offshoots in states of excitation, and that they preface and co-operate in the coagulation. In thrombo-plastic excitation, proceeding from the surroundings (the slide, particles of dust, or injured cellular elements) the thrombocyte sends out a protoplasm offshoot, which can adhere to other cells or to the slide. Agglutinates are formed in this way. Thrombokinase is given off from the spot where thrombocytes or agglutinates adhere to the slide, with the formation of thrombin and precipitation of fibrin as a result. Thus, the thrombocytes take

an active part in the coagulation. Fonio has also studied the appearance of kaolin granules in the formation of fibrin, and shows that the granules lie there as foreign bodies, and do not participate in any way in the process.

von Seemen and Binswanger (1928) have investigated the agglutination tendency of the thrombocytes before and after operations. They used Bürker's method, which is described as follows: A drop of blood is allowed to fall on a block of paraffin, to prevent coagulation. After a period of time the thrombocytes have collected in the upper part of the drop, which is then touched with a cover glass to remove a drop containing thrombocytes without admixture of other blood corpuscles. Before the operation, the microscope only reveals isolated thrombocytes and conglomeration of 2—3 thrombocytes, but on the 3rd—6th day after operation each agglutinate is found to consist most commonly of clumped and often numerous thrombocytes. There is no data as to how many cases were investigated and how often the samples were taken. Nor are any other figures given, so that the value of the statement bearing on the post-operative agglutination increase is difficult to assess.

Another author to take up this problem is Heusser (1928). He points out that Aynod found it was possible to use pig serum to produce separate agglutination without concurrent coagulation of the thrombocytes in a thrombocyte suspension present in citrated plasma. Guided by this, the author produced citrated plasma containing thrombocytes by centrifuging for half an hour, and this plasma contained numerous, non-clumped thrombocytes. To this plasma he added pig serum in different dilutions, shook the mixture and put it in a thermostat at 37° C for an hour. The microscopic examination of a drop after this time showed that agglutination occurs even in strong dilution of serum, contrary to the case before the operation. The method described by Heusser is open to the objection that the age of the serum is not given. Older serum loses its thrombin action, so that it is not impossible that it may undergo other alterations, too. There is consequently the risk of a source of error unless serum of the same age is used at the different occasions, or unless the serum taken has been preserved in a way to preclude changes of the kind mentioned. Moreover, there are no data as to the diluting fluid for the serum. Solutions for dilution other than Ringer's solution may occasion variations in the method's results

which cannot be controlled. In 10 investigated cases, Heusser has found that 7 cases presented isolated conglomerations with a serum dilution of 1: 4 before the operation, while the same solution on the 4th day afterwards gave great agglutinative tendency in 1 case, medium-sized conglomerations in 3 cases, and isolated ones in 6 cases. Since, as we have seen, the method may suffer from sources of error, and since nothing is said about the normal variation and the divergence between the values before and after the operation is small, it is difficult to form an opinion on the merits of Heusser's investigations.

Syller (1928– 1929) re-examined 50 cases with Heusser's method; he reports that increased conglutination of the thrombocytes can be observed postoperatively, and that this increase runs parallel with the rise in the sedimentation rate. There are no figures, so that the investigation cannot be assessed.

Windfeld (1933), who has made investigations with Heusser's method, is sceptical about it. He submits 10 cases with normal sedimentation rate investigated with this method, and shows that there is already agglutination on dilution of the serum addition 1: 16; this method is therefore hardly fit for investigating operation cases. Moreover, says Windfeld, all the agglutinates showed changed cellular forms on comparison with citrate-suspended thrombocytes without additional serum. He also maintains that if a sample is left to stand under the microscope, the thrombocytes agglutinate more and more; mere preservation can make a strong agglutination of a moderate one.

The above-discussed changes in the number of the thrombocytes, and the variations in the agglutination tendency which have been shown to be associated with operations and partus, have also led investigators to look for changes pathognomonic for thrombosis.

Kristenson (1928) has advanced that the thrombus should, by virtue of its structure, give thrombopenia in the blood while it is being formed, and that when further thrombocytes are added, an even more pronounced decrease in the thrombocyte count should occur. He supports this argument by a calculation based on the fact that a white thrombus of a certain size takes up so large a number of thrombocytes that this should affect the total number. Kristenson also describes a number of thrombosis cases where the thrombocyte count was calculated, eliciting a certain amount of proof for his theoretical speculation. However, as he himself stresses in his final

words, the number of cases must be looked upon as too small to allow of definite conclusions.

Normann (1928) has also interested himself in this question; he describes 4 thrombosis cases, in which the thrombocyte count was studied both before and after the operation. Normann states that these cases presented an abnormal postoperative thrombocyte curve. Either the increase to be expected was not forthcoming, or else there might even be a decrease in the thrombocyte count. An inspection of each case separately shows that, in one of the operations complicated by thrombosis, the thrombocytes rose from 207 000 to 300 000, i. e. an increase of 93 000. Of 8 normal cases with the same operation, 4 showed a smaller increase of the thrombocyte count, and 1 case had a slightly larger one, i. e. 95 000. The value of the submitted figures in thrombosis must be appraised with caution, the more so because there are only 4 cases with this complaint. Normann describes a further 7 cases, but these latter cases have no preoperative determination of the thrombocyte count, which means that they are no sure guide to a determination of the changes of the thrombocyte count in thrombosis. To illustrate the importance of the preoperative determination, it may be mentioned that, in the 4 cases of thrombosis where this was done, the values varied between 207 000 and 506 000.

Krukenberg (1930) reports that if thrombophlebitis or thrombosis occur in the puerperium, the thrombocyte count is low, in contrast to the rise normally setting in. He reports that low values could be established in 17 cases complicated by thrombosis. As, however, only two cases are illustrated and no figures given anyway, it is difficult to assess Krukenberg's investigation.

Windfeld (1933) has, however, several cases in his material where the thrombocyte count does not become larger or smaller than the original value, and where there are, nevertheless, no signs of thrombosis. He therefore considers himself to be dealing in these cases with a deficient power of regeneration in the thrombocytes. We may add that thrombosis may be present without manifesting clinical symptoms; the few cases which Windfeld advances cannot, therefore, be taken to confute the view of Normann and Kristenson.

The investigations carried out argue, then, that Kristenson's theory is correct, but there is no conclusive proof that this is so. However, as a decrease in the thrombocyte count can hardly manifest itself before a thrombus of a certain magnitude has been formed, the

method is scarcely practicable for early diagnosis of individual cases.

Jürgens (1939) considers that changes in the thrombocyte count do not affect thrombosis at all. He maintains that thrombi are never seen in thrombocytic crises in thrombopenics. This can hardly be used to refute the above-mentioned thrombocyte decrease associated with thrombosis, however, since in the one case—Kristenson and others—we are concerned with thrombopenia as a result of consumption, in Jürgens' case, on the other hand, with thrombopenia as a result of lowered production. Jürgens points out that there are normal thrombocyte counts in a number of thrombopathies, but that powers of agglutination are lacking. Jürgens and Naumann (quoted from Jürgens) claim to have shown that thrombocytes in a hanging drop of blood from a thrombopenic patient show deficient agglutination capacity. These circumstances lead Jürgens to consider the agglutination tendency of the thrombocytes to be more important than their number.

The agglutination of the thrombocytes at a certain point may be due to their having been placed there by conditions in the blood stream, and to their all having a tendency to agglutinate, but it may equally well be due to an emanation of agglutination-furthering substances from this point in the wall. However, Jürgens has studied the agglutination of the thrombocytes in some detail, and maintains that this phenomenon does not take place in protein-free suspension whatever substance he used in attempts to induce clumping (bacteria toxins, tissue juices, chemical substances). If protein substances are present they will undergo a change before or simultaneously with the agglutination of the thrombocytes. Supported by these findings, Jürgens considers the thrombosis to originate in a primary plasma change, while the agglutination of the thrombocytes is a secondary occurrence. The observations made were used as basis for a test worked out by Jürgens together with Schäfer and Klostermann. This test builds on the fact that ricin, from the seed of the castor oil plant, in a protein-free medium produces a thrombocyte agglutination, which is inhibited or maintained by qualitatively different protein bodies. The investigators mentioned found that normal serum has a stronger inhibitory effect than serum from thrombosis patients. The test is described as follows: To citrated plasma containing thrombocytes is added ricin in physiological salt solution. Inspection under

the microscope then shows a swifter agglutination in thrombosis cases than in healthy ones. The test is difficult to interpret. An equally plausible explanation would be to imagine that the citrate action is inhibited by the addition of ricin, and that what happens is only a coagulation of the fibrinogen present in the citrated plasma, caused by the thrombin factor present at the same time. What cannot be denied, however, is that there is a divergence in the relation of protein bodies in healthy cases and thrombosis patients. The cases investigated are few, however—a circumstance which Jürgens himself points out.

Summing up, it can be said of the *morphological conditions* associated with operations and partus that the variations are not specific to operations. Changes in the number of the erythrocytes and of the leukocytes may be due to circumstances unconnected with surgical intervention. However, the changes which it has been possible to establish do not seem to be very extensive. The leukocytes increase in number as a result both of the narcosis and of the operation. Their adhesion tendency seems to be raised in connection with partus, as also after an operation, though in this latter case the question cannot be considered definitively answered.

The number of the thrombocytes is raised after both operation and partus. Several authors have maintained that there are changes in the agglutination capacity, but these data cannot be assessed with certainty owing to inadequate methods and insufficient material for investigation in the different series.

The literature on these matters is scanty, and even more meagre is that dealing with morphological blood changes and thrombosis. It has not yet been possible to demonstrate any established change which bears a relation to thrombosis.

An extensive literature from the early part of the 20th century deals with the question of the *plasma changes* following operation and also thrombo-embolism. In the first place the changes occurring in the plasma proteins, and consequently also in the sedimentation rate of the erythrocytes, were studied. The refractometric and viscosimetric methods used by most of the authors proved to be not very reliable, however, and the results need not, therefore, be considered here. This applies to most of the works of Reiss (1902, 1913), Ro-

bertson (1910, 1915), Naegeli (1931), Windfeld (1933) and many others. Furthermore it has been shown by the studies following Fåhræus's (1918) pioneer work on the sedimentation reaction that morbid conditions are almost invariably followed by changes in the composition of the plasma proteins. Thus, it is not surprising to find that several authors claim to have shown that there is an increase in the globulin: albumin ratio, an increased fibrinogen content, and an increased sedimentation rate after surgical operations. The same thing is also to be expected in thrombo-embolism, with its increase in temperature and its local influence upon the blood vessels and the tissues.

Variations in the sedimentation rate after operation have attracted great interest, but it would be too great a digression to review everything that has been written on this question here. We will only touch on the results submitted by Öwre (1929) and Windfeld. The former has investigated a large material with Westergren's method. He shows that the previously known rise after operation does exist, and that it depends on the magnitude of the operation, as also on complications (infection, haematoma). As a rule, the sedimentation rate rises to its maximum on the 3rd—4th day, to return to normal on the 8th—10th day in younger people, and a week or so later in older ones. Öwre is unable to find any relation between thrombosis and sedimentation rate, but states that if the former does set in, it causes a rise in the latter; obviously, then, there is a relation, though it is not pathognomonic.

Using the same method Windfeld, too, has studied the influence of the operation on the sedimentation rate; like Öwre, he found the latter to show a typical rise, with its maximum on the 3rd—4th day. The sedimentation rate yields no typical result for different operations, and Windfeld is unable to establish any influence from age.

Summing up, it can be said that a decrease in the total protein obviously sets in after operation. The globulin: albumin ratio undergoes shifts, with increase of the globulins, a circumstance which has been demonstrated by all methods employed. There is a rise in the viscosity after operations. The fibrinogen content in the blood also increases and will be discussed in greater detail in another connection. The sedimentation rate rises after operation up to the 3rd —4th day, afterwards slowly falling to normal. It has not been possible to establish any relation between total protein, globulin

increase or rise in viscosity, on the one hand, and thrombosis on the other. Nor do the sedimentation rate and thrombosis seem to have any connection with one another.

Since Virchow's time, opinions as to the importance of the *blood coagulation* for the formation of thrombi have varied between two extremes. On the one hand, thrombosis has been looked upon as a coagulation, on the other, thrombosis and coagulation have been considered essentially different processes. Thus, during the end of the 19th century and first decades of the 20th century, thrombosis and coagulation were taken to be two separate conceptions; during the last decade, however, the idea of thrombosis as a process closely akin to blood coagulation has come more and more to the fore. This has meant that the substances active in this reaction have become of vital interest, and have been subjected to very thorough studies. Increased content of these substances in the blood has been connected up with thrombosis and looked upon as a predisposing factor; attempts have also been made to find a relation between thrombosis and special changes in the concentration of these substances. This interest has been greatly stimulated by the possibility of favourably influencing thrombo-embolism with ordinary blood constituents such as heparin.

It is not possible to discuss now the copious literature on questions belonging here. We will only affirm that, as the inhibition or reduction of the blood's coagulation tendency can prevent the formation of thrombi or arrest their development, it is reasonable to assume a relation between the coagulation mechanism and thrombosis. It is at present impossible to determine whether this latter is a true intravasal coagulation, but it can hardly be wrong to assume that the substances reacting in coagulation influence the formation of thrombi. A certain amount of light should be obtained from an inspection of the literature dealing with coagulation. Here, as previously, one is referred to works dealing with postoperative and puerperal changes in the composition of the blood. Before this literature comes under discussion, something should be said about the prevalent view of the coagulation mechanism of the blood.

The theory as to what happens when the blood coagulates was first formed by Schmidt (1892), and developed by works of Pekelharing (1892), Hammarsten (1899) and, later, Spiro and Fuld (1904).

Morawitz (1905) united the results of the researches in this field into a whole, the so-called classical coagulation theory, on which our view of the coagulation process is still based. According to Morawitz, this process has two phases: (I) the coagulation ferment thrombin is formed from the preliminary prothrombin stage by the action of thrombokinase in the presence of calcium ions, after which (II) the fibrinogen is converted into the insoluble fibrin under the action of thrombin, probably through denaturation. A number of other theories have been advanced, but only the one mentioned above has stood the test of critical examination, and at the same time been possible to link up with more recent discoveries.

Prothrombin is the inactive precursor of thrombin. The amount of prothrombin is determined after its conversion to thrombin, by measuring the effect of the latter on a fibrinogen solution. In so doing, however, all prothrombin must have been transformed into thrombin for correct values to be obtained. Building on this circumstance Quick (1935 –36) has elaborated a method for calculating the prothrombin content in plasma. Using a temperature of 37° C with optimal recalcification of oxalate plasma and surplus of thrombokinase, he turned all the prothrombin present into thrombin. which, as soon as it forms, exerts its effect on the plasma fibrinogen. He does not work with a known fibrinogen solution, quite simply measuring the coagulation time of the plasma. The method gives fairly constant values although, theoretically, it is not faultless. Warner, Brinkhous and Smith (1936) raise the following justifiable objection to it, namely, that two phases are included in it, both of which may vary separately; these are the time taken for the thrombin to form, and the time the fibrinogen needs to turn into fibrin. Besides this, the fibrinogen content may vary. These authors have therefore worked out a method whereby these two phases are separated from each other. Though there is thus an error in Quick's method, its use has led to results which seem to show that the content of prothrombin may vary in connection with pathological processes engaging the liver. Thordarson (1941) has submitted a method where the oxalate plasma is replaced by a known fibrinogen solution. and Lehmann (1941) has modified Quick's method applied to small quantities of blood.

Thrombokinase, or thromboplastin, is the activator of prothrombin, according to the classical coagulation theory. Its true nature is still unknown, however. The cephalin fraction of the phosphatides has a strong thromboplastic action. A still stronger activity is ascribed to certain thermolabile lipoproteins (Chargaff, Ziff and Moore, 1941). Thrombokinase has a widespread distribution in the tissues of the body, and is thought to occur in a bound form in the blood — more exactly, in the corpuscular elements, above all the thrombocytes. The amount of thrombokinase bears a definite relation to the formation of thrombin, as Eagle (1935) has shown. The amount of thrombin formed is, within certain limits, always the same, regardless of the amount of thrombokinase; only the time factor is influenced, an increased quantity of thrombokinase being accompanied by a swifter formation of thrombin. Eagle has also shown that a thrombocyte suspension, added in varying quantities to a prothrombin solution, activates a certain amount of prothrombin, but that the time factor is influenced by changes in the number of thrombocytes.

Hammarsten has, in several investigations, submitted proofs of the necessity of *calcium* in the first phase of coagulation in physiological systems. In a work of 1896 he makes the following pronouncement: 'The presence of calcium is only necessary for the formation of the coagulation ferment thrombin, not for the effect of the finished thrombin on its substrate fibrinogen, i.e. for the actual formation of fibrin.' I shall not here go into the obscure question of how calcium acts in the formation of thrombin, but will only say that different authors have been able to establish that variation in the calcium content causes variation in the coagulation. As early an investigator as Hammarsten (1899) reports that a concentration of 0.05—0.200 % calcium has a favourable effect on the formation of fibrin in a system of purified thrombin and fibrinogen, whereas 0.625% inhibits it. Fleig and Lefébure (1902) state that, in whole blood, the coagulation may even be prevented with 2 % calcium chloride. The optimum content for coagulation is given at 1.15 %. Mellanby (1909), too, has found that the variations in the calcium content set up changes in the coagulation time. It is not yet clear whether the calcium content also influences the transformation of prothrombin into thrombin, seeing that the calcium content in mammalian blood

is fairly constant. This factor shows very little, if any, relation to the coagulation rate of ordinary blood.

As has already been indicated, Eagle was able to show that the amount of *thrombin* formed was the same, regardless of the amount of thrombokinase. He claimed to have found the amount of thrombin to be proportional to the prothrombin content. Being an enzyme, the thrombin can be quantitatively determined only by measuring the coagulation time of plasma or a fibrinogen solution. Wöhlisch (1929) states that the thrombin concentration is in inverse proportion to the coagulation time, a view which is also shared by Quick (1936). However, other authors, including Eagle (1935), have been able to find inconsistencies in this view. It is hardly necessary to go into the literature on these questions; suffice it to say that an increase of the thrombin content shortens the coagulation time.

Eagle reports that a quantity of 0.003 cm^3 of purified thrombin solution causes fibrinogen to coagulate in 20 minutes, while 1 minute is required if 0.2 cm^3 of the solution is used. He does not say how many analyses he made, but the difference is so great that the figures may well be considered conclusive.

As to the influence of the content of *fibrinogen*, Mellanby (1909) reports that a rising content of fibrinogen inhibits the coagulation, while Foà (1913) and Yamada (1918) have reached a completely opposite view. None of these authors gives the magnitude of the fibrinogen content. In his work of 1933, Schmitz intimates that a concentration varying between 0.04 and 0.22 % does not have any influence on the time factor in the coagulation process. Using investigations in which the fibrinogen content in the coagulation system varied between 0.0275 and 1.6 %, Wöhlisch, Diebold and Kiderlen (1936) have shown that there was a coagulation optimum in the fibrinogen content between 0.3 and 0.4 %. Higher concentrations had an inhibitory influence. These results should explain the contradictions in the investigations quoted above.

The effect of thrombin on the fibrinogen is influenced by the prevailing temperature, but different authors have different ideas on this question. Mellanby (1909) states that, in the stage between 5° and 18° C, a rising temperature increases the rate of coagulation, while temperature variations between 18° and 52° C set up no change.

Rettger (1909) submits a somewhat smaller range of temperature to cover a constant rate of coagulation, namely between 17° and 41° C. Stromberg (1911) shares this latter opinion. Landsberg (1913) reports that the coagulation time in systems of $MgSO_4$ plasma and serum, from plasma + activated serum, and of fibrinogen + thrombin, show lower values with rising temperature up to 37° C. He also maintains that the temperature must have a double effect in coagulations where a ferment is active, since higher temperature on the one hand accelerates fermentation, and on the other destroys or inactivates it. These two effects are concurrent, thereby influencing the reaction. This may explain why the action within a certain range of temperature remains fairly constant. Using a system of $MgSO_4$ plasma and thrombin solution produced according to Schmidt, the last mentioned authors were able to find that temperatures between 18° and 30° C had no influence on the coagulation time. For pure fibrinogen solution and pure thrombin solution the optimal value for the coagulation rate lies between 37° and 40° C. The same also holds for fresh serum and $MgSO_4$ plasma. Yamada, Kugelmass (1923) and Kuvashima (1923) submit about the same temperature limits; their works do not give the fibrinogen percentage, however, which seems to vary considerably. Stromberg, for example, like Landsberg, uses diluted $MgSO_4$ plasma; Rettger, on the other hand, takes a pure fibrinogen solution obtained from oxalate plasma by precipitation. This means that the investigations of these authors cannot be compared as they stand. Wöhlisch, Diebold and Kiderlen (1936) have shown that, when the fibrinogen content shows values of less than 0.2 %, the effect of the temperature decreases as the concentration gets less. A content of about 0.14 % fibrinogen is coagulated by the same amount of thrombin as quickly at 20° as at 37° C. A later experiment with another thrombin preparation shows that the influence of the temperature only ceases at a concentration of 0.06 % fibrinogen.

Summing up, it may be said that the influence of the temperature varies with changes in the fibrinogen content. Swifter coagulation results from higher temperatures up to 34° C. There is no change in the coagulation time in the region between 34° and 40° C, provided there is a content of at least 0.08 % fibrinogen. When plasma is used for calculating the thrombin effect, it may not be diluted below this minimum content of fibrinogen; moreover, the temperature

must be kept constant, and be the same for each determination. These measures are necessary if the different determinations are to be comparable.

The inhibitory action of *heparin* on the coagulation of the blood, together with its presence in tissues and blood, caused even Howell to look upon it as the natural anticoagulant of the body, and to consider it to be the substance which *in vivo* prevented the intravasal coagulation of the blood. Although this opinion could by no means be considered proved, it quite naturally concentrated attention on the possibility of preventing thrombosis by increasing the heparin content of the blood. A whole series of investigators have envisaged the prospect of thus solving one of the great problems of medicine. However, many attempts to this end were frustrated by the fact that pure heparin was not to be had.

Any detailed account of the literature dealing with heparin is outside the scope of the present thesis. Excellent monographs on this topic are now available (Mason, 1939; Jorpes, 1939; Astrup, 1944).

As soon as pure heparin became available through the work of the Toronto laboratory (Charles and Scott, 1933 and 1936) and of Jorpes (1935), the substance was used in order to counteract and prevent thrombo-embolism. As a result of these experiments it became evident that thrombotic processes could be checked both experimentally in animals and in thrombosis in man. In a series of papers the Toronto workers showed that induced experimental thrombi in animals could be prevented by means of heparinization (Murray, Jaques, Perret and Best, 1936; *ibid.* 1937; Best, Cowan and MacLean, 1938). In Sweden, interest was concentrated on the possibility of preventing postoperative thrombosis by heparinization during some period after operation (Crafoord, 1937; Bergquist, 1940; Wetterdahl, 1941, 1943; Leissner, 1941, 1942). The powerful influence of heparin on the coagulation mechanism—an influence which is even strong enough to prevent thrombotization of the vessels—is hereby amply demonstrated.

According to Morawitz (1905), the spontaneous decrease of coagulatory activity in serum, the *antithrombin effect*, is due to the transformation of the thrombin into a inactive stage, which he calls metathrombin. However, he demonstrated a coagulation-inhibiting

substance, which he called antithrombin, in serum and plasma also a substance which as old an investigator as Schmidt (1892) assumed to be present in the circulating blood. This view was verified by several authors, Mellanby (1909) for one, who showed that serum had an antithrombin effect, and that small quantities of serum could inactivate considerably larger quantities of thrombin. Howell and co-workers (1911—1918), in particular, studied the antithrombin effect of serum and found it to be due to a thermolabile factor which had its greatest effect at 37° C. Tissue extract counteracted the antithrombin effect. A further finding of Morawitz was verified, namely that antithrombin requires a certain incubation time to take full effect. The antithrombin effect was at a quite early date thought to be due to the coupling of the thrombin with a protein (Rettger, 1909). This idea has subsequently been accepted by a number of authors; others rejected it, but the question was not really clarified before the discovery of heparin, which stimulated closer studies of these circumstances. To begin with, the issues were confused by the introduction of this new factor into the discussion, but it gradually became clear that two separate substances, independent of one another, were in question. Quick (1938) considers the antithrombin effect to depend on a particular fraction of the serum albumin. He showed that the coagulatory capacity of thrombin which had been in contact with serum albumin for 1 minute was lowered to one-hundredth part of its original activity, while a contact with serum globulin for 30 minutes only lowered the coagulation effect by about half. Comparing the different degrees of speed with which serum and serum albumin neutralized thrombin, Quick was furthermore able to show that the antithrombin effect depends on the albumin content in the serum. He considers the antithrombin effect is specific for serum albumin, and cannot be explained by simple absorption.

There has been a certain difference of opinion as to how the antithrombin and heparin stand to one another. Heparin is thermostabile and has not the character of a protein, unlike antithrombin, which is thermolabile and belongs to the albumin group. Heparin is present in very small quantities in the blood, cf. Charles and Scott (1933) and Wilander (1938); in a pure state it does not have antithrombic character (Howell), but acquires this property if it is introduced into the blood channel or to blood *in vitro*. Various explanations to this phenomenon have been sought, but the prevailing

opinion nowadays seems to be that the heparin does not acquire its full effect until it is bound to a CO factor (Brinkhous and collaborators 1939) which, according to Quick (1938), and Astrup and Darling (1941), is an albumin. Astrup and Darling (1942) describe a method they have worked out for determining the antithrombin effect. Their method has been used by Volkert (1942) for studies bearing on the antithrombin content of the blood, its effect and how it stands to heparin. However, Volkert's investigations are of no interest to this present thesis apart from his finding that antithrombin is latent in the blood. According to him, its action depends on two components. One of these, constituting about 80 %, is a fairly constant substance, whose effect can be influenced by loss of blood but not by Indian ink injections. The other normally constitutes about 20 %, but may be appreciably increased during pathological states. This latter component can be inhibited by the injection of Indian ink, and its formation can be checked by the same measure. It shows a certain similarity to the effect of heparin coupled with the CO factor, while the nature of the first component is not clear.

The above-mentioned transformation of thrombin into metathrombin has been shown by works of Weymouth (1933), Gasser (1917) and Rich (1917) to be an antithrombotic effect in its own serum; consequently, metathrombin would seem to be a combination of thrombin and antithrombin.

Influence of Operation and Childbirth

Investigations into the changes of the *thrombokinase* content have been impossible, owing to insurmountable difficulties in determining the quantity of this substance in the blood. The discussion on its bound form in the blood has not led to clarifying results, and no definite knowledge of the state of affairs here has been reached. Moreover, as thrombokinase is a catalyst, its amount only influences the speed at which thrombin is formed, and not, as indicated above, the quantity formed. If a sufficient amount of thrombokinase is introduced into the vascular system an intravasal coagulation occurs; this has been shown by numerous experiments carried out by Takaura (1937), Lenggenhager (1941), and others. It has not been

possible to show whether thrombokinase appears in the blood in connection with operations, but it seems probable that it does so. Tissue lesion liberates thrombokinase: tissue is injured in operations: consequently, thrombokinase is thereupon formed.

The literature on the changes in the *prothrombin content* is extremely copious, but the investigations into its postoperative variations are few. Those that exist deal mainly with changes in the prothrombin content associated with icterus with cholemic haemorrhages. Nevertheless, some authors have studied non-icteric cases, also.

Smith, Ziffren, Owen and Hoffman (1939) show with graphs how an operation (cholecystogastrostomy) for obstructive jaundice is followed by immediate decrease of the prothrombin content. In connection with Vitamin K treatment of surgical cases with icterus, Butt, Snell and Osterberg (1939) describe postoperative decrease of the prothrombin value, but these authors do not give any figures, either. Without describing his investigations in any detail, Butt (1939) reports that no changes in the prothrombin content occur in operations where the liver is not touched, or where its function is not disturbed. Stewart (1939) submits a postoperative decrease of 20 to 40 % in the prothrombin content of gall patients with icterus. However, he gives no figures for the number of cases investigated or for the prothrombin values, so that this work, too, cannot be appraised. Tage-Hansen (1940, cit. from Borgström) describes 4 cases of icterus where he was unable to find any postoperative fall in the prothrombin content. On the other hand, Strömbeck (1941) found a postoperative decrease on the first day after operation in 6 cases of choledochus affection, of which 3 were, however, not icteric. As the material in both cases is small, no definite conclusions can, of course, be drawn. Garrot-Allen and Livingstone (1941) give an account of 97 patients, who were operated upon under different anaesthetics for different complaints, none of which involved the liver or biliary tract. There was no variation in the prothrombin time before and after the operation. Operations for obstructive jaundice and biliary fistula were in 15 cases followed by a fall in the prothrombin. The divergence of the figures is so great that statistical calculation would be superfluous.

Borgström (1943) describes a material from the Surgical Clinic

of Lund in a study into the prothrombin content in operations not involving the liver or the biliary tract. This material consists of 101 patients operated upon for various complaints—none of them icterus, however. His material, which is statistically calculated, shows no significant differences before and after operation.

The literature has thus shown that, with ordinary methods of analysis, no postoperative divergences in the prothrombin content are to be demonstrated where the operations do not involve the liver.

Brinkhous, Warner and Smith (1937) have studied the prothrombin content in pregnant women; these investigators were unable to find any divergences on comparison with normal conditions. They give no figures, however, so that the full force of the report cannot be assessed. However, Thordarson (1941) has used a method worked out by himself to investigate a relatively large material (112 pregnant women, and 25 puerperants); he showed there to be a rise in the prothrombin content, with its maximum in the 9th month. After partus, the prothrombin sinks to normal; this occurs in the 3rd—4th week. The differences are so great that, although there are no figures which allow of statistical calculation, Thordarson's statements may well be taken as conclusive. The same author has submitted analogous figures in his doctor's thesis (1941). He maintains that there was no complication with thrombosis in his material, despite high prothrombin values in the puerperium.

The lack of unanimity among different authors is probably due to their having used different methods. Thordarson's method gives a variation round the mean of 100, the lowest being 85 and the highest 115, and with 6.58 as the calculated standard error of the mean value. The method error has not been calculated. Smith, Warner and Brinkhous use their own method, but since—as mentioned above—their divergence from Thordarson is not supported by figures, no appraisal can be made of their data.

An investigation by Marx and Bayerle (1943) shows guineapigs suffering from scurvy to have a prolonged prothrombin time, which fell to normal on treatment with Vitamin C. The coagulation time for whole blood was normal, while the capacity of a thinned $MgSO_4$ plasma to be coagulated by thrombin (standardized strength) was usually found to be increased in severe forms of scurvy. On calculation, the figures show that the data can be verified statistically.

The normal value of the coagulation time (20 cases) is 132 ± 5 seconds; in animals with severe scurvy (14 cases) it is 77 ± 4 seconds. The divergence, 55 ± 6.4 seconds, is thus significant.

In studies bearing on the postoperative variations in the *calcium content*, Allen (1927) claimed to have found a decrease after 24 hours, and yet another after 3 days. An inspection of his graphically presented curves hardly substantiates this statement. The course of a number of them shows slight variations, while some show a rise on the very day when others show low values. Obviously, the question cannot be assessed without statistical calculation of the material, and as there are no primary figures, this is impossible.

A number of other authors, such as Öwre (1929), Windfeld (1933) and others, have interested themselves in this question. However, the materials seem to be incomplete, and such variations as exist are small, so that these works are not reviewed in any detail.

The *thrombin content* in the blood can only be calculated indirectly, on account of the labile nature of the thrombin. By experiments on animals whose pancreatic duct had been ligated, Wohlgemuth (1917) showed with a method worked out by himself in 1910 that the fibrinogen content in the blood increases greatly on the 1st—3rd day after the operation, while the thrombin values are constant. The method is pretty rough, however, and can hardly give the exact values desirable. No figures are available.

Studies into postoperative changes in the thrombin content have been made by Boshamer (1927), who reports that the thrombin value and the antithrombin content go hand-in-hand in that a high thrombin content causes high antithrombin values. The investigations are only summarily described, however, so that no definite significance can be attached to them. Boshamer has also investigated the amount of thrombin and of antithrombin in association with operations and reports that, in these conditions, there is an increase of the thrombin content and a decrease of the antithrombin action. This disturbance of the thrombin-antithrombin equilibrium increased up the the 5th 7th day after the operation, afterwards slowly returning to normal. Boshamer illustrates his findings with graphs (18 cases) but gives no primary figures.

Öwre (1929), too, has investigated 5 operation cases, using a varia-

tion of Wohlgemuth's method. He states that the content of thrombin may vary in different individuals, and that no effect from the operation is demonstrable. However, his material is too small to clarify the situation.

Lenggenhager (1936) discusses in an essay the 'Frühgift' after operations, which is dealt with here and there in more recent literature. It is thought to be a toxic substance occurring in freshly defibrinated blood and in press juice from muscle. Lenggenhager points out that the toxin in question is taken by several investigators to be of importance for thrombosis, since they consider it able to lower the blood pressure and set up endothelial alteration and thrombocytic agglutination. König (1934) attributed the toxicity to the content of degenerative products from cell nuclei. Lenggenhager points out how strange it is that a supposed toxic action should decrease very rapidly, to have disappeared after about 2 hours. He has shown that rabbits die very soon after intravenous injection of muscle press juice or defibrinated blood, even when this latter is from the same species. Heating to 60° C removes its effect. Press juice from boiled muscle is also innocuous. Now, the point is that thrombin is also destroyed when heated to 60° C, and the effect of injecting press juice and defibrinated blood can be neutralized by previous administration of heparin or hirudin. On the strength of these investigations and argument, Lenggenhager claims to have shown that the so-called 'Frühgift' is nothing other than thrombin.

Investigations linking up thrombosis and increase of thrombin content are of relatively early date. For example, in his studies on eclampsia gravidarum, Dienst (1902) found that thrombi were strikingly often revealed in autopsies on infants dying in association with the partus of an eclamptic mother, or on women who had died from eclampsia. He sets this in relation to the blood change which he considers to be present in the diseases mentioned, and stresses that both the blood of the mother and that of the child shows raised fibrinogen concentration. Dienst also states that the fibrin ferment (thrombin) is increased, but his work provides no support for this statement.

In a later work (1908) Dienst has again asserted that the increased quantity of fibrinogen and the increased number of leukocytes, above all degenerating leukocytes, in eclamptics are the cause of the thromboses which may arise, and which are often demonstrated in these

patients. Here, the degenerating leukocytes are taken to be purveyors of thrombin, which is formed in the blood of the mother and then passes over to the child, for the thrombi shown in the child are, according to Dienst, of later date than those of the mother. Dienst supports this statement by the following experiment. Using a rabbit, he induced a sterile pleura exudate, which he removed after the animal had been killed. The exudate was divided into 2 portions, one of which was divested of leukocytes (by filtration through a bacteria filter) immediately, the other not until it had been kept for 24 hours in a cold-storage room, and had been stirred repeatedly with a rod. Dienst assumed that the leukocytes gave off their contents to the fluid during this procedure. Injections of these different filtrates into doubly ligated ear veins of rabbits now showed him that the first filtrate was not capable of inducing coagulation, but that the second was. To infer from this experiment that the fibrin ferment is derived from the leukocytes is not compatible with the present-day view. However, degenerating leukocytes give off thrombokinase, which acts on the prothrombin and turns it into thrombin; despite the reservations made here, therefore, Dienst's argument supports his opinion. Backed up by theoretical arguments. Dienst further maintains that the outflow of the fibrin ferment into the blood of eclamptics is the crucial point in the thrombosis which can be demonstrated in them. All the same, he is unable to advance any direct proof of this opinion of his.

In yet another work (1920) Dienst develops the view, indicated here, that an increased content of fibrinogen (above all of thrombin) in the blood is the cause of eclampsia, and therewith of the thrombosis arising in connection with it. This time the proofs he offers are firstly a number of cases of eclampsia whom he saw recover after being given hirudin intravenously, and secondly the fact that the blood of 5 cases of eclampsia investigated by him showed a shorter coagulation time than that of normally pregnant women. Neither proof is completely satisfactory, however, since the number of investigated cases in both is too small to be considered conclusive. Dienst has also performed experiments on animals. He injected fibrinogen and fibrin ferment into rabbits, obtaining thromboses and also changes in liver and kidneys, resembling those appearing in eclampsia.

However, Dienst states that pregnant women with kidney lesion, and above all eclamptic patients, show an increased thrombin con-

tent in the plasma; this he claims to have shown with Alexander Schmidt's method of producing thrombin. He gives no figures or details, however, so that this statement, too, is not to be evaluated. Dienst also discusses antithrombin variations and thrombin changes in the plasma and serum of pregnant and eclamptic cases respectively. In so doing, he bases his statements on coagulation experiments with different dilutions of serum and plasma from the blood of eclamptic women, which experiments are compared with similarly performed experiments on blood from healthy women and from normal gravidae. There are consequently no data as to the method's normal values or as to variations from individual to individual, so that its exactness cannot be estimated. Dienst nevertheless comes to the conclusion that there is more thrombin, and therefore less antithrombin, in the blood of pregnant women than in that of non-pregnant women, and that this difference is even further accentuated between eclampsia cases and healthy, non-pregnant cases.

This theory of Dienst's is open to the comment that, if fibrin ferment really does exist free in the blood of the pregnant woman thrombosis would always be present to a greater or lesser degree. However, Dienst assumes that the thrombin which is formed is balanced by the antithrombin of the liver, and that surplus thrombin is secreted by the kidneys. Lesions in the liver and kidneys upset the ability of these organs to neutralize the effect of the thrombin, which therefore develops to excess, with thrombosis as one of its consequences. It is, of course, conceivable that thrombin arises in the blood in this way, but it seems a more plausible theory that eclamptic patients and, to a lesser degree, normal gravidae show an increase of prothrombin (cf. Thordarson), and that a swifter formation of thrombin is brought about by an increased quantity of cells and their degeneration products. It should be reasonable to assume that the intravasal formation of thrombi is favoured in the presence of increased prothrombin and increased possibilities of providing thrombokinase.

Lenggenhager (1941) has also studied the part played by thrombin in thrombosis. He says that thrombokinase is liberated at operations by the lesion to the tissue, and that this kinase is resorbed via the lymph channels. When the thrombokinase has reached the blood, it is given the chance to react with the prothrombin and transforms it into thrombin. This thrombin turns some of the fibrinogen into

fibrin, which adheres to the thrombocytes on account of its affinity for them. The thrombocytes are thereby rendered liable to agglutination, and the aggregates thus arising may, if the blood stream is slow, adhere to the vascular wall and form thrombi. The thrombin is also bound to the fibrin and therefore to the thrombocytes, and it is not given off into the blood until later, when it has become inactive metathrombin.

Lenggenhager states that persons liable to or threatened by thrombosis have greater difficulty in transforming thrombin into metathrombin. In support of this statement he carries out the following reaction. 30 cm^3 of blood are taken from the cubital vein by means of a puncture; half is mixed with citrate solution, while the other half is defibrinated by mechanical whisking with a glass rod. The different portions of blood are centrifuged, to given non-corpuscular citrated plasma and serum respectively. One part serum, one part plasma and several drops of 5 % $CaCl_2$ solution are mixed together at a fixed temperature (22° C), and the coagulation time is measured. After half-an-hour the procedure is repeated. Sera which break down the thrombin poorly give a strong thrombin effect in both the first and the second sample (short coagulation time), while normal sera yield large variations between the half-hour and one-hour values. Lenggenhager claims to have found that sera in thrombo-embolism at an early stage or even preoperatively always show a poor breaking-down effect, i. e. a slight difference between the half-hour and hour values. However, as he gives no figures, it is impossible to assess his statement. In any case, the method has no uniform test solution to test the sera against. The citrated plasma can, of course, be accepted as test solution, but only if it has been shown by experiment series that there is no difference between the citrated plasma of the investigated subject and a uniform citrated plasma, which is the same at each investigation. In an experiment of this kind, the prothrombin in the citrated plasma must also be expected to be changed into thrombin at the recalcification, and be added to the serum thrombin. It might furthermore be mentioned that the calcium addition, 5 drops of $CaCl_2$ solution, seems to be arbitrarily chosen (cf. Lehmann). Moreover, the movement effect should be constant for, as Feigel (cit. from Wöhlich) has shown, an inconstant effect may affect the amount of the thrombin formed. Finally, we might mention that, though the breaking-down effect is admittedly measured by this

method, it may be influenced by a variation both in the amount of thrombin and the antithrombin content. A reaction always takes up a certain amount of time. If the investigated blood is assumed to contain so much thrombin that only a part of this thrombin is needed to reach the minimum time for the coagulation, so great an effect must remain, in spite of the breaking down, that the minimum time can be arrived at even after a further half-hour.

Lenggenhager's investigations have been retested, giving rise to a variety of opinions. Thus, in a work of 1942, Stortz has described 16 cases of thrombosis and embolism or phlebitis, showing low values for Lenggenhager's reaction. Yet it should be mentioned that one case with thrombi in both calves showed a normal half-hour value. Furthermore, Stortz does not say how long after the diagnosis of the thrombus the determination was made. This investigator is more cautious in his pronouncements than Lenggenhager, and considers that results are not obtained with this reaction until a thrombus is formed.

Manstein (1943), who completely accepts Lenggenhager's statements, has found a number of cases presenting low values; he considers that thrombosis must have been present in these cases, although it could not be demonstrated by current clinical methods. In 8 cases of 13 women with unquestionable thrombosis, the reaction was strongly positive, while in 4 the values were not quite so distinct. No figures are given, so that no appraisal can be made of the statements advanced. However, Manstein gives an account of a further 28 cases, 19 of which showed no signs of thrombosis despite low values for the reaction, while the remaining 9 showed signs of embolism. Thus, this material does not seem to have provided Lenggenhager's reaction with that support which Manstein claims to have shown.

Schneider and Kickelhayn (1943) have investigated 260 cases, but were unable to verify Lenggenhager's statement that low preoperative values are to be regarded as disposing to thrombosis. These authors quote an as yet unpublished work by v. Kaulla and Olivier; of these authors' 140 cases, 46 showed values which, according to Lenggenhager, would prove thrombosis, but there was no thrombosis. All these authors have also found a fall in the values on the 3rd and 5th day after operation. A circumstance arguing far more forcefully against Lenggenhager's view is, however, that thrombosis occurred in 5 of 152 cases with normal values. Kaae (1943) has also made a

re-investigation, and shown that there may be low values preoperatively without thrombosis setting in and that, conversely, thrombosis occurred in 2 cases even though they had shown high values before operation. Concerning the investigations touched on here, it may be mentioned that thromboses were often not diagnosed. It not infrequently happens that a fatal embolism sets in just when the patient is about to get up after an operation followed by no complications. It therefore seems plausible that a low value is combined with a thrombosis without its being possible to establish the latter's existence. Investigations by Schneider and Kickelhayn, Kaulla and Olivier, and Kaae seem, however, to have shown Lenggenhager to be wrong in saying that low preoperative values are conducive to thrombosis. Nevertheless, despite his small material, Stortz has shown that manifest thromboses may possibly be attended by a low value for Lenggenhager's reaction.

Thus, opinions are divided as to the thrombin content after operation. As none of the different opinions are supported by substantiating figures, this question must as yet be considered unsettled. As regards the thrombin inactivation which is determined by Lenggenhager's method, it seems as though it presented a decrease after operation. Theoretically, however, the method is not completely trustworthy, so that this question, too, must be said to be still unsolved. A decrease in the breaking down may very well be attributed to the presence of a larger quantity of thrombin. Lenggenhager's statement that disposition to thrombosis can be gauged by the reaction he described has been shown by re-examinations to be incorrect.

The task of the *antithrombin* in the blood has been taken to be the counteracting of the intravasal coagulation. Changes in the concentration of this substance may therefore be thought to bear a relation to thrombosis. As early an investigator as Howell (cit. from Nielsen) reports that two thrombosis cases showed a lower antithrombin value than the one that is customary in normal individuals. Denny and Minot (cit. from Nielsen) confirm this finding. Boshamer (1927) reports that, in one case of thrombosis, the antithrombin curve deviated from the usual course in that it returned rapidly to normal after the operation; this he took to indicate that the increased thrombin content had an opportunity to develop its effect. Boshamer submits 9 cases of thrombosis, investigated immediately, and 13 cases in-

vestigated 1—2 days after the thrombus had been established. He reports a strong antithrombin decrease in these cases. Some figures are mentioned, but as the normal value is not given, their significance cannot be assessed.

Against these statements we have the results of Nielsen's investigation (1943). This investigator, who used Astrup and Darling's method for determining the antithrombin value, reports determinations carried out on 6 patients at 15 minute intervals during the operation, on 12 patients daily up to the 9th day, and on 6 patients with thrombo-embolism; in no case was there any change in the antithrombin value. The diverging results may well be due to different methods of determination. However, the nature of the antithrombin effect has not as yet been fully elucidated, so that the works mentioned here are difficult to assess. The situation as regards antithrombin during operations and in association with thrombosis must be looked upon as still to be cleared up.

As it would seem from the above-mentioned works that the substances reacting in the coagulation are subjected to changes in connection with operation and partus, it is also probable that the *coagulation time* of the blood shows changes. A number of authors have maintained that this is so, while others reject such a view. The possibility of a relation between a change in the coagulation time and the occurrence of thrombosis has also been discussed. Some authors claim to have found such a relation, whereas others claim the opposite.

Hartmann (1909) has found a shortened coagulation time after operation, and Adler (1912) reports the same situation after partus. Rabinovich (1926) maintains that, in 100 cases operated under ether, 80 showed a shortening of the coagulation time during and after the operation. Bültemann (1929) submits a material of 500 cases and states that the capacity to coagulate rises on the 1st day after operation, to return slowly to normal on the 4th—5th day. Goosens and Liebich (1939) have reached a similar view. Their material, which consists of 20 cases, has been investigated immediately before and immediately after operation, and a calculation of their results gives the means of 550 $\pm$ 102 seconds and 201 $\pm$ 24 seconds respectively. The divergence of 349 $\pm$ 105 seconds is consequently significant. Starup (1942) describes a material of 200 cases with investigations before, during and after operation. He reports that, already

on the day of operation, there are lower preoperative values than were present a few days previously. In the 75 patients which Starup investigated during the actual operation, the samples have been calculated at intervals of 5—10 minutes. The figures elicited are plotted on graphs, and the curve shows a steep fall as early as 15 minutes after the beginning of the operation, at which level it remains until the end, afterwards gradually rising. Different operations are reported as reducing the coagulation time to different extents; large operations, especially intraperitoneal ones, reduce it most. The time the operation takes is not assigned any influence. Starup has later on followed the postoperative coagulation time in 146 cases every hour up to 10 hours, and thereafter daily until the 10th day. He reports it to rise slowly to normal values, which are reached during the 2nd —4th day after the operation. Starup has not submitted any primary figures. He has only given the degree of the changes, and sometimes he merely illustrates the results by graphs. Despite these deficiencies, which make it somewhat difficult to assess the results of the investigation, the material and the differences are of such a size that the findings made can probably be taken to represent the actual state of affairs.

A number of authors hold the view that the coagulation time does not undergo any change, and that no irregular changes appear in connection with operation, pregnancy or partus. Neu and Kreiss (1911) report that no variations in the coagulation time are to be detected during pregnancy, at partus or in the puerperium, and Keller (1912) was not able to find any difference, in 43 cases, between the preoperative value and the values determined immediately after operation or later. Chauvin, Esmenard and Jaur (1926) have arrived at the same view. They submit figures from 25 cases. A calculation of these values gives the mean of 16.7 ± 0.4 minutes before the operation, and 15.8 ± 0.5 minutes after the operation; that is to say, the difference (0.9 ± 0.6 minutes) is not significant. Allen (1927) found no divergences in 8 cases; the means, calculated approximately from the curves, were 99 seconds before the operation, and 74 seconds on the 6th day after the operation. In view of the smallness of the material, however, the difference of 35 seconds is probably too little to be considered conclusive — a fact which Allen himself points out. Öwre (1929) has arrived at the view that changes in the coagulation time are associated with operations, but are not uniform. He has

investigated 24 cases and not found that they followed any law. A calculation of the figures supports this view. Before the operation the mean is 130 ± 7 seconds, and after the operation (1st day, 20 determinations) 108 ± 8, making a divergence of 22 ± 10.6 seconds.

After an investigation comprising 51 cases, Windfeld (1933) arrives at the same opinion as Öwre. Both use the same method of determination. The samples were determined 1—2 days before the operation, and then every day after it, in some cases up to 20 days. A calculation of the values before the operation (30 determinations) gives a mean of 125 ± 4 seconds; the mean on the 2nd day after the operation (26 determinations) is 112 ± 4.7 seconds. There is thus no significant difference (divergence = 13 ± 6.2 seconds). On the 6th day after the operation (12 determinations) the mean is 98 ± 6 seconds; i. e. the divergence is 27 ± 7 seconds. In view of the scanty material, this difference can hardly be considered significant, though there is a certain likelihood of its being so.

The contradictory data in the literature may be due to the use of different methods, but may also be due to differences in the treatment. Besides this, several authors give no exact times, as to when in relation to the operation the determination is made, and this may also have played a part. Moreover, the coagulation time may be affected by different drugs, as also by loss of blood. It is furthermore possible that variations in the coagulation time may occur at different times during the 24 hours, as also that infections or other pathological states of the organism at operation may exercise an influence.

Investigations into some of these factors have been carried out a long time ago. Thus, in 1905, Sahli (cit. from Wöhlisch) described shortened coagulation time after blood losses. By means of experimental studies on man with venesection and calculation of the coagulation time,von den Velden (1909) claims to have shown that the loss of blood lowered the coagulation time to one half or even one quarter the normal figure. McClintock and Magers (1927) have also studied these matters, and report an increased coagulation tendency after loss of blood experimentally induced in dogs. If the animals first underwent splenectomy, the curtailing of the coagulation time could not be observed after venesection. Vosburgh and Richards (1903) have pointed out that adrenalin causes an increased coagulation tendency, and in a work by Cannon and Gray (1914) the authors maintain that excitation of the splanchnic nerves has the same effect:

this is attributed to the fact that excitation of these nerves sets up an increased production of adrenalin. Different authors, e. g. Whipple and Hurwitz (1911), Cannon and Gray (1914), Mendenhall (1914), maintain that other drugs, such as chloroform, chloral hydrate and ether, also affect the coagulation time. In these works, however, the primary figures have been illustrated by graphs, so that the investigations are difficult to assess. Nevertheless, as the variations from the normal values seem to be conclusive, it should be possible to say that at all events adrenalin has been shown to shorten the coagulation time in small doses, and to prolong it in large ones. As regards ether, it has been shown that narcosis with this drug decreases the coagulation time, and that this decrease is further accentuated if adrenalin is given simultaneously.

Zopf (1932) reports that, when dogs are under deep ether narcosis, there is a swift and pronounced fall in the coagulation time, which does not regain normal values until the 5th—6th day after the operation. There are no primary figures, so that any appraisal of these data is impossible. H. Geissendörfer (1935) states that ether narcosis causes an increased coagulation tendency, which is also enhanced by operation, and does not return to normal until the 3rd—4th day. Geissendörfer, too, gives neither the number of cases nor any primary figures; he illustrates his findings with graphs, and this makes it impossible to assess his work. Holm Nielsen (1942) has investigated the influence of different circumstances on the coagulation time, and states that venesection (500 cm^3) brings it down to one quarter the normal value within 15 minutes. This author was able to show that the injection of adrenalin increased the coagulation tendency, and the same effect was observed after metrazol shock. However, all his investigations are illustrated solely by graphs, which makes a calculation of the material impossible. Nevertheless, as his curves show great fluctuations in their courses, his statements may well be probable.

As so many different circumstances influence the coagulation time, great difficulties are encountered in studying it. It should therefore be hard to try and find a connection between *thrombosis and changes in the coagulation time* at operation or the days immediately following. A number of authors have nevertheless directed their attention to this question, and submitted a number of findings.

Thus, Küster (1913) states that there is no connection between thrombosis and coagulation time. Bülteman (1929) submits thrombosis cases (numbers not given) where the coagulation time did not, as he had found, return to normal after operations, low values persisting until the 8th day. The absence of figures detracts from the worth of the report.

Starup (1943) maintains that a strong increase of the coagulation tendency bears no relation to thrombosis. He backs up this statement by saying that strumectomies, for example, have a pronounced fall in the coagulation time during the operation, and that such cases are usually free from thrombosis. He does not support this view with any figures, however, and his statement is in any case not proved, since he gives no account of the changes in coagulation time associated with thrombosis. It may also be pointed out that a marked fall in the coagulation time during the operation need not be connected with thrombosis, since the coagulation time comes under the influence of a number of different circumstances during that period. Of primary interest in this connection is the time when thromboses usually manifest themselves.

The investigations of latter years into how far blood changes are present at thrombosis can be summed up as follows. As thrombosis is a common complication after operations, and as it often occurs during the puerperium, also, the line adopted has been to investigate blood changes postoperatively and after partus. Such changes are interesting in themselves, but even so the fact that an operation upsets the concentration or effect of the different components of the blood does not necessarily have any bearing on the formation of thrombi. Added to this, the methods at our disposal for determining the constituents of the blood are not absolutely exact; moreover, changes may occur at random. The differences which are to be observed after an operation in a small number of cases do not in themselves prove connection with the operation; they may in the first place indicate a random change. They may furthermore have to do with recumbency, or with other factors attendant on surgical intervention. In these circumstances it is not surprising that the results obtained have not been of a more definitive character, and have consequently not led either to a final solution of the

thrombosis problem or even to a starting-point for further investigations along definite lines.

Changes in the number of leukocytes have been observed in association with operations. In themselves, these changes are of no very great interest, since they might also be expected to be due to the diet given, or to possible infections. For that matter, the differences observed are hardly of a more striking character. The adhesion tendency of the leukocytes shows a moderate increase in connection with partus, and the postoperative course also shows an increase for several days. These latter changes are not particularly great, however.

The thrombocytes have been found to show an increase after operation and partus. Their agglutination tendency seems to be connected with their surroundings, and therefore follows the plasma changes. No definite proof of a relation to thrombosis has been possible.

The total protein seems to decrease after operation. The protein substances in the plasma show an increase of the globulins, which has been demonstrated both with direct determination and indirectly from the sedimentation rate. This latter shows rising values after operation, and runs parallel with an increase in the fibrinogen content. It is plain that the blood changes discussed here are on the whole the same as those observed in infectious conditions of the organism. It is therefore hardly likely that there is any specific connection between blood changes of this kind, on the one hand, and thrombosis on the other. Nor has it been possible to establish any such connection.

The different substances participating in coagulation may show changes in connection with operations. Opinions seem to be divided as regards a number of these substances, however. Thombokinase is generally considered to appear free and in greater quantity in association with operations. The prothrombin content is unchanged in operations not involving the liver and biliary tract. Operations involving this region are attended by hypoprothrombinaemia, particularly if the patient has icterus. The prothrombin increases during pregnancy up to partus, and the puerperium also shows an increased prothrombin content, which gradually returns to normal, however. The calcium content does not seem to be affected by operations, and it has not been possible to show any connection with thrombosis.

According to the literature the thrombin content in the blood seems able to change in connection with operations, and it has also been maintained that these changes play a part in the formation of thrombi; there are no proofs for either of these contentions, however. Changes in the conversion of the thrombin to an inactive phase also seem to exist after operations, but no clarity has been reached either as to this question or as to the possible relation of these changes to thrombosis. The same is true of antithrombin and its action in connection with operations. The fibrinogen is increased during operations and after partus, as has already been indicated above.

It would seem as though the coagulation time is shortened in connection with operations, although the question cannot be said to be fully cleared up. This is due partly to the fact that several factors influence the time for the coagulation of the blood, but is also to be ascribed to the lack of uniformity attached to the methods of investigation. A number of authors have not described their methods, others have used such as may make for uncontrollable influences on the coagulation time. As a rule, no figures are given, so that statistical calculation is out of the question.

According to some authors, there is a relation between shortening of the coagulation time and thrombosis; others deny any such relation. There are no investigations with sufficient material, so that the question is still to be solved.

As most of the different substances investigated in connection with operations touch one another at certain points or have an effect on the coagulation, the investigations into the coagulation time after an operation constitute, in a way, a verification of the investigations made into the different constituents of the blood. On the whole, these investigations have not yielded any striking results, either. However, there seems to be a decrease of the coagulation time immediately following an operation, which is not surprising, since loss of blood, adrenalin, excitation of the splanchnic nerves and ether narcosis enhance the tendency of the blood to coagulate.

The data as to the connection between blood changes and thrombosis are still scantier. We have not yet got as far as being able to state that a certain blood change is a fairly trustworthy forerunner of thrombosis. Indeed, it has not yet been possible to prove that certain blood changes are either pathognomonic for thrombosis or

incompatible with this disease state. However, certain investigations rather point to a connection between thrombosis and a number of blood changes. Thus, it has been shown that the adhesion value of the leukocytes is raised in the postoperative course during those days when thrombosis usually sets in. Furthermore, it has been maintained that both increased thrombin content and shortened coagulation time for whole blood bear a relation to thrombosis — a view which, as thrombosis and coagulation seem to be kindred processes, is probable, though by no means proved. An investigation on a larger material would probably give, if not a final solution of the problem, at least an answer to certain unclarified questions which are linked up with it.

As appears from the summary given here, no clarity has been reached as to the adhesion tendency of the leukocytes at operation and in association with thrombosis. Opinions differ as to how the coagulation tendency of the blood behaves in connection with operations and childbirth, and in thrombosis. Different authors hold divergent opinions on the question of the amount of thrombin, but there seems to be no investigation into the thrombin effect present at the very time of the operation, childbirth and thrombosis. Nor does anything seem to have been published on the behaviour of the mast cells in operations. It is these problems that the author has sought to investigate more fully in the work presented here.

CHAPTER II

Material

The material for the present investigation has been collected in different hospitals during the years 1940—1944. In view of the nature of the investigation—it includes, for example, blood tests of several different kinds—it has been necessary for practical reasons to collect different materials for the various sections in the work. Thus, the clinical material for determining the coagulation time in whole blood has been taken from the Surgical Department of the Lidköping hospital; the material for calculating the adhesion tendency of the leukocytes and the content of mast cells in the blood comes from the Surgical Department of the Västerås hospital, and the material for calculating the thrombin effect has been taken from the Surgical Departments of the Karolinska hospital, Academic hospital in Uppsala and the Örebro hospital respectively, from the Köping hospital and, finally, from the Maternity Department at the Arboga hospital.

The material used to study the effect of an operation on the mast cells consists of 37 cases: 17 men and 20 women, aged 30—60 years. The material has been chosen to include those operations which are attended by great lesion to the tissue, and which are relatively often accompanied by thrombo-embolism. The material is admittedly small, but it is large enough for the projected calculation, and should be representative as, apart from the nature of the operation, no particular requirements had to be observed when collecting.

The material for studying the adhesion tendency of the leukocytes comprises 163 cases, 63 men and 100 women. The age distribution is outlined in table 4.

Table 4

Age	15–19	20–24	25–29	30–34	35–39	40–44	45–49	50–54	55–59	60–64	65–69	70–74	75–79
No. of cases	2	8	17	16	12	28	23	14	18	7	12	5	1

Table 5 shows the distribution of the material according to the operations.

Table 5

	Minor operations, local anaesthetic	Major operations, spinal anaesthetic or general narcosis				Infected cases
Nature of operation	Hernia ingu. Hydrocele Varicocele Vasectomy Prolaps. ut.	Stomach op.	Cholelithotomy without infection	Uterus + adnexa op.	Cancer op.	Appendicitis acuta Cholecystitis acuta or chronica
No. of cases	34	49	23	27	14	16

When possible, more serious operations have been chosen, in view of the greater liability to thrombosis complications. Otherwise the material has been selected at random, so that it can be considered representative. This material contains 7 cases of thrombosis.

The material collected for an investigation of the variations in the coagulation time associated with operation and partus consists of 173 operation cases and 28 partus cases, the former comprising 108 women and 65 men. The age distribution is shown in table 6.

Table 6

Age	5–9	10–14	15–19	20–24	25–29	30–34	35–39	40–44	45–49	50–54	55–59	60–64	65–69	70–74	75–79	80–84
No. of cases	1	0	3	3	9	12	23	23	23	22	15	14	16	5	3	1

The distribution according to operation is as follows (table 7):

Table 7

	Minor operations, local anaesthetic	Major operations, spinal anaesthetic or general narcosis				
Nature of operation	Hernia ingu. Hydrocele Varicocele Vasectomy Prolaps. ut.	Stomach op.	Cholelithotomy without infection	Uterus + adnexa op.	Prostate op.	Cancer op. of the stomach and intestines
No. of cases	30	28	47	36	15	17

Thus, the material consists mainly of major operations, these being chosen when possible, in order to obtain cases with thrombo-embolism. This material contains 15 cases of certain thrombo-embolism, in addition to those given above.

The material for investigating the thrombin effect consists of 82 partus cases and 72 operation cases. The partus material has been assembled without selection, all cases admitted to the department being made the object of investigation. Some of them were excluded, however, as a result of lack of time or of difficulties in taking samples. The material is thus representative. The tables below (8 and 9) show the distribution according to age and to number of deliveries respectively.

Table 8

Age	15–19	20–24	25–29	30–34	35–39	40–44
No. of cases	6	19	22	25	8	2

Table 9

Age	I-para	II-p.	III-p.	IV-p.	X-p.	XII-p.	Total
No. of cases	42	22	10	6	1	1	82

The method followed in collecting the operation material has been to take mainly more serious operations from the Karolinska hospital, whereas in the departments of the other hospitals all operations were investigated, except those where a difficulty in obtaining samples (deep-lying blood vessels) proved an obstacle. The material comprises 72 cases, 22 women and 50 men. Table 10 shows the distribution of the material according to age.

Table 10

Age	15–19	20–24	25–29	30–34	35–39	40–44	45–49	50–54	55–59	60–64	65–69	70–74	75–79	80–84
No. of cases	2	6	3	7	4	3	13	5	12	8	5	2	1	1

The distribution according to operation is as follows (table 11):

Table 11

	Minor operations, local anaesthetic	Major operations, spinal anaesthetic or general narcosis				
Nature of operation	Hernia ingu. Vasectomy Prolaps. ut. Laesia menisk. Appendectomy Hydrocele	Stomach. op.	Cholelithotomy without infection	Uterus + adnexa op.	Prostate op.	Cancer op. of the stomach and the intestines
No. of cases	19	9	14	7	10	13

This material contains 17 cases of certain thrombo-embolism in addition to those given above. The material could not be larger because nowadays every suspected thrombo-embolism is immediately treated with heparin or dikumarin and therefore can not be investigated for the thrombin effect. However, the material is large enough with regard to the tallying figures and can, after statistical treatment, provide answers to the problem in question.

CHAPTER III

Methods

The material in the present work has been investigated with current clinical methods. These investigations were part of the hospital routine, with determination of haemoglobin, sedimentation reaction (s. r.), counting of red and white corpuscles, etc.

In the special investigations made by the present author, the number of white blood corpuscles was determined with the usual method, i. e. staining with ordinary dye fluid and counting the cells in Bürker's counting chamber. Further, *mast cells* were stained and calculated according to Holmgren's method (1941), i. e. by making a smear specimen in the ordinary way, letting it dry in the air and fixing it in 1 % solution of toluidine blue and 60 % alcohol for at least 20 minutes. The specimen was then rinsed with distilled water for some seconds. The mast cells appeared in this specimen with large, red granules and the nuclei resembling clover-leaves. The number of mast cells per 400 white blood corpuscles was determined. The mean of three such calculations (i. e. of 1200 white corpuscles) expressed in per cent gives the mast cell value. The method error was calculated from 18 double determinations on 18 healthy subjects. The first blood sample was taken from the tip of one of the fingers of the right hand, the second from another finger. The method error was calculated as 0.079, which may be denoted large in view of the fact that the means of the pathological material differ only slightly from the normal value. For this reason it is hardly possible to obtain a diagnosis from single determinations; this will be discussed later. The normal value of the mast cells was determined with values from 50 healthy conscripts (50 single determinations) and gave the mean 0.181 $\pm$ 0.019 %, with a dispersion of $\pm$ 0.137 %; the method error 0.079 therefore amounts to 43.6 % of the normal value.

The *adhesion tendency of the leukocytes* was determined by v. Philipsborn's method, which he describes as follows. The finger-tip is first carefully cleansed with ether; from it is then taken one blood sample for counting the leukocytes, and one sample up to line 6 in a pipette intended for sample-taking in blood corpuscle counts. This latter sample, which must be sucked up as quickly as possible, is blown out on to a slide made of ground non-alkaline glass. This slide has been warmed in a thermostat at 37° C, and after receiving the drop it is placed in a similarly warmed moist chamber and then kept in a thermostat at 37° for half an hour. After this time, the drop is rinsed off the slide with distilled water, the slide is left to become air-dry and is stained with diluted Giemsa solution. Usually, the adhering leukocytes are now evenly distributed over the specimen, but if this is not the case v. Philipsborn takes the places where they are densest and counts the number of adhering cells in 100 small squares, with a Zeiss micrometer fitted into a Zeiss ocular, and divides the sum by 10. Two counts are made, and the ratio of their mean to 1000 leukocytes in the blood is taken. This method has been somewhat modified by the present author, however, since it was found that the rinsing should be done with Ringer's solution, the hypotonic water greatly damaging the leukocytes and thereby rendering an exact count impossible (see fig. 1). Moreover, to resort on occasion to the places with the greatest agglomeration of cells seemed far too arbitrary a proceeding. Consequently, the calculations were not made on special areas; instead 3 areas selected at random were taken. The adhering corpuscles consisted mainly of leukocytes, with an occasional lymphocyte (see fig. 2).

The method error was calculated from 19 double determinations on 19 healthy subjects, and was ± 0.429 or, in per cent of the normal value, 7.7 %. The normal value of the adhesion value was calculated from 50 healthy conscripts and gave the mean 5.6 ± 0.05, with a dispersion of 0.366.

As appears from the literature on the subjects, the reason for the migration of the leukocytes to the surface of the glass has not been clarified. Nor is there as yet a solution to the problem of the actual adhesion. The migration of the leukocytes may be due to their amoeboid movement, which during pathological states in the organism increases or diminishes, depending on changes in the medium (cf. Vejlen's experiments with hypertonic NaCl solutions);

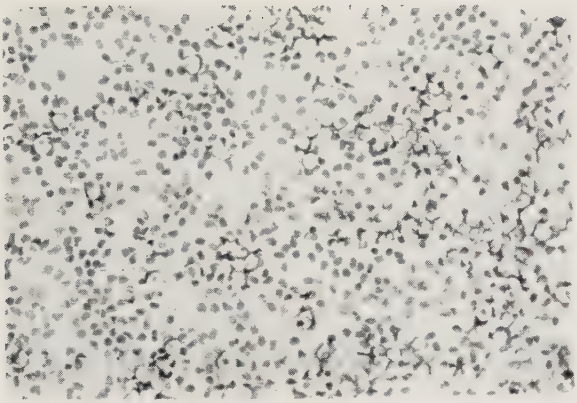

Fig. 1 a

Fig. 1 b

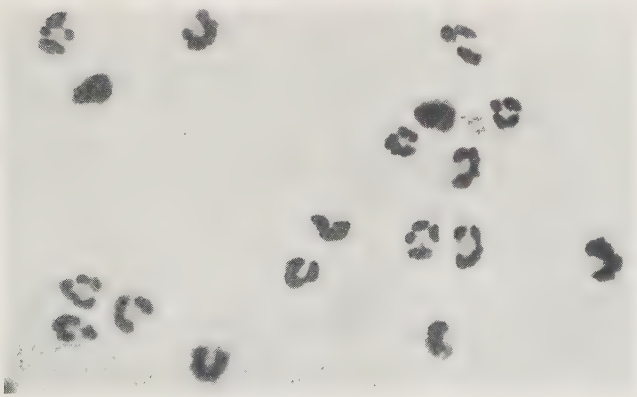

Fig. 2

Fig. 1 a. Specimen rinsed with H_2O.
Fig. 1 b. Specimen rinsed with Ringer's solution.
Fig. 2. Specimen showing adhered leukocytes and also lymphocytes.

another possible explanation is that surface tension forces act on the leukocytes to a greater or lesser degree according to their sensitivity under different conditions. As regards the actual adhesion, this may be thought to be a tendency to adhere which proceeds from the leukocytes and which also is linked up with the amoeboid movement (cf. Vejlen's experiment), or it may be an interfacial phenomenon between glass and fluid.

We shall here describe an experiment which, though it cannot claim to elucidate the problem, nevertheless seems to show that interfacial forces of glass and fluid exert an influence on the migration of the leukocytes to the surface of the glass. The adhesion values was calculated on 50 healthy conscripts, both with specimens made in the customary manner and with a hanging drop. The figures are given in table 12.

Table 12

Adhesion of leukocytes. Normal value

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Ordinary specimen			Hanging drop		
n	$M \pm \varepsilon(M)$	σ	n	$M \pm \varepsilon(M)$	σ
50	5.624 $\pm$ 0.052	0.366	50	5.532 $\pm$ 0.097	0.686

The difference of 0.092 $\pm$ 0.11 thus shows there to be good agreement between the two values. A special influence from the surface of the glass or from the interface between glass and fluid must therefore be presumed to exist, as otherwise diverging values should have been obtained. That an interfacial phenomenon between glass and fluid, and not a simple interfacial effect, e. g. between air and water, is involved is shown by the fact that if we take a smear specimen from the cupola of the drop in accordance with the sketch in fig. 3, we only get a few red corpuscles when using an ordinary specimen; there are absolutely no white corpuscles. If a smear preparation is made from the cupola of the hanging drop (in both cases at the juncture

Fig. 3.

when the drop is to be rinsed away), it yields an abundance of red blood corpuscles and 1—2 white corpuscles per visual field. The reason for this is, of course, that more corpuscles sink towards the cupola of the drop when it is hanging, in obedience to the laws of gravity. A number of white corpuscles which do not adhere to the slide will then be collected here.

There are several methods for determining the coagulation time in whole blood. The present author has used Petrén's method; though in a modified form, for practical reasons. The change made is that the blood is taken from the finger-tip, i. e. capillary blood is used instead of venous blood. Furthermore, a definite quantity of blood is used, measured with a pipette, and finally, the temperature is kept constant in different determinations.

For this method an apparatus (see fig. 4) has been constructed with thick walls of glass and wood. Inside the apparatus there is a watch glass running in grooves, one of which is movable, so that the glass can be rolled backwards and forwards by alternately pulling and pushing the movable groove. The thick walls ensure that the temperature can be kept practically constant (22°) during the coagulation. A definite quantity of blood (blood sucked up to 7th line in a pipette used for taking samples in leokocytes counts; carefully blown out on to the watch glass down to line 1) can be poured on to the watch glass through a hole (9), which can be stopped up with a cork. The blood is taken after puncturing a finger-tip cleansed with ether. The blood is left to well out without stasis, and the first drop is wiped away; it is the next drop that is used. The time is taken from the moment the drop is sucked up into the pipette. During the first minutes the apparatus can be rotated round the movable groove, so that the drop of blood on the watch glass is kept in motion. After 2—3 minutes the apparatus is held up against a window, and the watch glass is rolled back and forth with the movable groove (1). The blood is examined against the light, and the time taken when clots show signs of forming (formations of fine threads in the thin layer of blood in the upper part of the drop).

A calculation of the normal value (morning value) for men and women, as also the value for different days, has been made, and the result is given in table 13. We find here that there are no differences between the sexes, and that the values for different days tally well. The two materials have therefore both been made into one for cal-

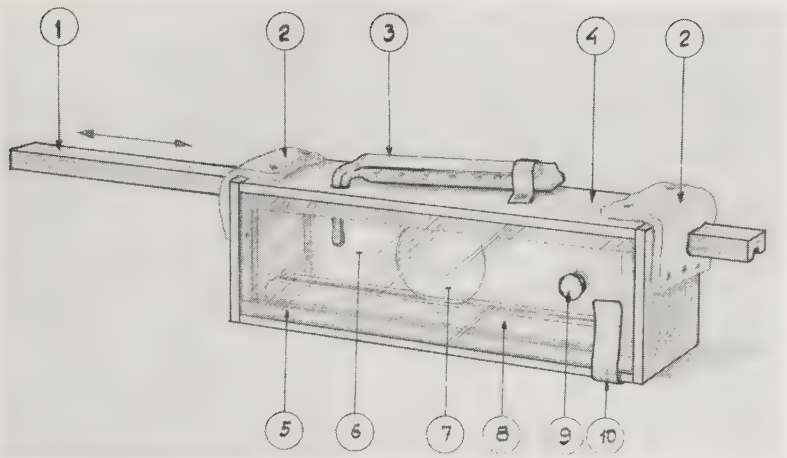

Fig. 4.

1. Movable wooden bar with longitudinal groove for watch glass. 2. Packing felt. 3. Thermometer. 4. Wooden case, hermetically sealed. 5. Packing rubber frame. 6. Sheet of glass, on each side of the case. 7. Watch glass. 8. Fixed wooden bar with groove for watch glass. 9. Rubber cork in the hole of one glass sheet. 10. U-shaped clip to keep the glass sheets in place.

Table 13

Coagulation time different days

$M \pm \varepsilon(M)$ = arithmetical mean ± standard error of the mean.
σ = standard deviation.

	I Men			II Women		
	Number	$M \pm \varepsilon(M)$	σ	Number	$M \pm \varepsilon(M)$	σ
1st day	24	395.6 ± 5.5	27.2	14	388.9 ± 8.1	30.4
2nd »	24	388.5 ± 4.8	23.7	14	385.6 ± 5.6	21.0
3rd »	24	392.4 ± 4.9	24.0	14	385.9 ± 6.4	24.1
4th »	24	390.4 ± 3.7	18.1	14	393.4 ± 6.4	24.1
5th »	23	395.4 ± 4.7	22.6	14	388.4 ± 5.6	21.1

Diff. (I–II) = + 6.7 ± 9.8
+ 2.9 ± 7.4
+ 6.5 ± 8.1
— 3.0 ± 7.4
+ 7.0 ± 7.3

culating the normal value and gave the mean 391.6±3.3, with a dispersion of 20.2.

The calculation of the method error and the normal value was made on a material of 37 fracture cases who were out of bed; it was done at a juncture when the effect of the fracture on the blood could be presumed over (as a rule one month after the fracture), and when the sedimentation reaction was normal. The means for morning and evening show a slight difference, as can be seen from table 14.

Table 14

Coagulation time. Normal values

$M \pm \varepsilon(M)$ = arithmetical mean ± standard error of the mean.
σ = standard deviation.

Morning			Evening		
Number	$M \pm \varepsilon(M)$	σ	Number	$M \pm \varepsilon(M)$	σ
37	391.6 ± 3.3	20.2	37	392.7 ± 3.5	21.5

The difference is only 1.1 ± 4.8 seconds, which shows that there does not seem to be any difference in the coagulation time on the

morning and evening of the same day. This means that it should be legitimate to calculate the method error on two calculations made on the same day. This has been done, and the method error was 16 seconds or, expressed as a per cent of the normal value, 4.1 %.

To illustrate the importance of the quantity of blood when calculating the coagulation time, 20 cases have been calculated, blood to line 10 of the pipette being used in the one case, and to only line 3 in the other. The determinations were made at the same time on 3 consecutive days. The results are outlined in table 15.

Table 15

Coagulation time in different quantities of blood

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Line 3			Line 10		
Number	$M \pm \varepsilon(M)$	σ	Number	$M \pm \varepsilon(M)$	σ
20	287.3 ± 9.1	40.5	20	381.3 ± 6.8	30.2
20	288.5 ± 9.2	41.5	20	384.5 ± 7.0	31.4
20	289.0 ± 9.0	40.2	20	384.3 ± 6.4	28.4

As can be seen, the coagulation time is considerably shorter if the pipette is filled only to line 3. The value of the coagulation time varies only slightly from day to day.

The importance of always using the same drop to come from the finger-tip is plainly shown in the graph of values given below. Here, 16 healthy subjects were investigated every hour between 8 a. m. and 8 p. m., and every other hour between 8 p. m. and 8 a. m. The investigation also included calculations made with the blood taken both at once (the first test) and after 1 and 2 minutes respectively. A considerable curtailing of the coagulation time clearly follows the use of blood coming from the wound in the finger-tip after the lapse of one minute. This is probably due to an increased quantity of thrombokinase in this blood. Judging from this material (which is, however, too small to warrant definite conclusions), there would hardly seem to be any changes in the coagulation tendency during the 24 hours. A lower value may possibly be indicated between 12 noon and 2 p. m.

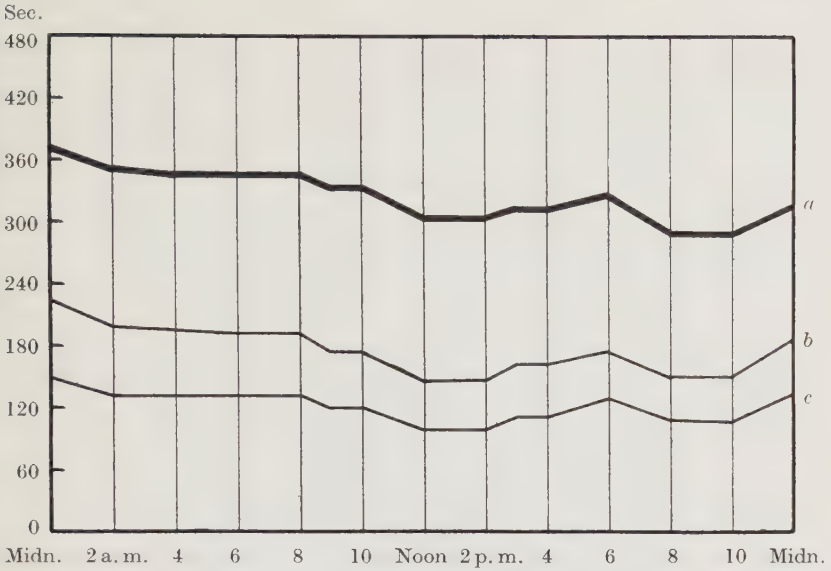

Fig. 5. Time of coagulation during the 24 hours.
a = 1st sample, b = sample after 1 min., c after 2 min.

A comparison between the method described by the present author and that of Hedenius—which has been analysed in respect of the method error by Sköld (1944)—shows the latter to yield the larger method error, i. e. $\pm$ 8.7 %, that of the present author being 4.1 %. A comparison between the normal material of Sköld and the one presented here shows the two to disagree as to the actual variability, i. e. that part of the total variability which is contingent on inter-individual variations. Sköld's normal material (women), where the dispersion is 1.7 minutes with a method error of 0.52 minutes, shows an actual variation of $\sqrt{1.7^{-2} - 0.52^{-2}} = 1.62$ minutes, while in the author's material, 20 determinations on 20 healthy women, the actual variability is $\sqrt{30^{-2} - 16^{-2}}$, i. e. 34 seconds. This difference in variability must be due to the fact that some part of the two materials compared is not representative. Possible explanations are either that some factor in the author's material had a stabilising effect on the coagulation tendency, or that some of the subjects in Sköld's material displayed abnormally prolonged (conductors?) or curtailed coagulation time. In this instance the author's material consisted of 20 healthy nurses and assistant nurses at Lidköping

Hospital. The meals, working conditions and room temperatures were the same in the entire material, nor were there any very great variations in age. The determinations were carried out in the same room, and within the course of a few hours. It is possible that these uniform external conditions had a stabilising influence on the coagulation tendency. It seems a more reasonable assumption, however, that Sköld's material contained cases with pathological values for the coagulation time. The question cannot be definitively settled, however.

To determine the thrombin effect, the author used a method elaborated by himself from Lenggenhager's 'Thrombinabbaureaktion'. When possible, blood without the addition of salts, tissue extract or the like should be used, to get an idea of the thrombin effect which may occur or be achieved. The thrombokinase (if any) in the blood adds its effect to that which may be exerted by the corpuscular elements. If ready-made thrombin is present—as may be expected after operation and partus—a co-operation with the thrombin formed later may be presupposed. As the amount of thrombokinase only affects the time factor, studies of thrombin must allow sufficient time for the transformation of prothrombin into thrombin. Further points to notice are that the thrombin effect decreases with keeping, and that this decrease in effect varies with varying temperatures. Heat accelerates it, cold slows it down. The reason for this is now assumed to be that the inactivation of the thrombin in native serum is due to a factor in the serum called antithrombin. This demands a certain time, not less than 15 minutes, to take full effect.

Taking these circumstances into account, the following procedure has been adopted to determine the thrombin effect. A slightly compressed vein in the fossa cubitalis is punctured, and about 6 cm^3 of blood sucked up in an ordinary dry record syringe with a short, wide and sharply ground cannula. Five cm^3 of this blood is emptied into a glass beaker, which is placed in an apparatus constructed by the author, called a defibrinator (see fig. 6). This consists of a stand (1), in which are placed a block (7) for the beaker and the movable electric motor (3) set in a frame. The motor drives a blade (5). When the motor is brought down towards the beaker, it sends the blade into the beaker; at the same time the current is switched on, so that the motor starts and the blade begins to rotate. The fibrinogen is thereby changed into fibrin and adheres to the blade. Seven-and-

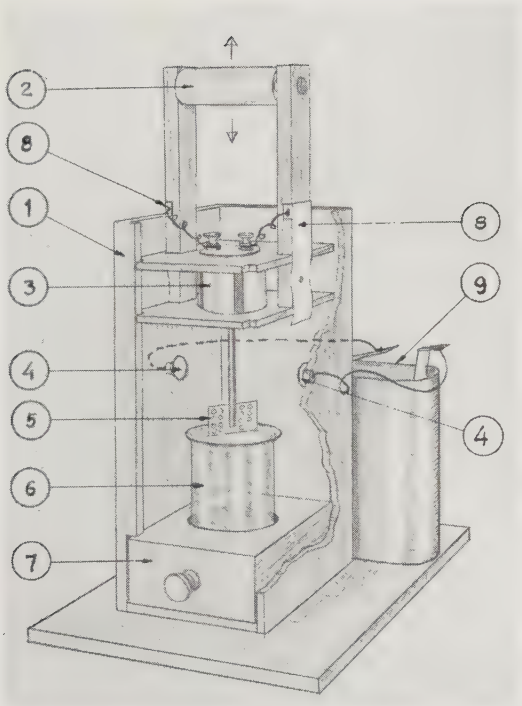

Fig. 6.

1. Wooden case. 2. Handle, movable vertically. 3. Motor. 4. Switch. 5. Stirrer. 6. Glass beaker. 7. Movable block. 8. Contact sheets. 9. Pocket-torch battery.

In the figure the handle and the motor are in their upper position. When the handle is pressed down, the stirrer is lowered into the beaker. At the same time the current is switched on and stirring starts.

a-half minutes after the blood has been sucked up into the syringe, the defibrination—which is now total—is interrupted, and the blood is centrifuged for 7 $^1/_2$ minutes. After centrifuging, the clear serum layer is pipetted off into a storage test tube, where it is left to stand for 15 minutes at 22°. After this time, 0.2 cm³ of citrated plasma (1 cm³ 3.7 % citrate solution + 4 cm³ blood centrifuged for 10 minutes) and 0.2 cm³ of serum are added together by means of a blood sugar pipette in an ordinary Ellermann tube. The tube is shaken gently without stopping, and a fine fibrin film gradually appears in the solution. The time in seconds taken for this coagulation is a gauge of the thrombin effect.

It is of no consequence to the value of the thrombin effect whether the blood sample is taken with or without stasis, or if the blood is made to run freely from the cannula into a glass beaker. Blood taken from one arm without stasis and from the other arm after stasis for 5 minutes gives values with no or only slight differences.

The test substance used to start with was uniform citrated plasma (from the author), and calculations (during the time 12 noon to 2 p. m.) made on 45 healthy men (75 determinations) and 32 healthy women aged about 30 years. The figures are given in table 16.

Table 16

Thrombin effect. Normal values

$M \pm \varepsilon(M)$ = arithmetical mean ± standard error of the mean.
σ = standard deviation.

Women			Men		
Number	$M \pm \varepsilon(M)$	σ	Number	$M \pm \varepsilon(M)$	σ
32	328.60 ± 30.83	174.38	75	287.47 ± 23.10	200.07

There is no significant difference between the two values (41.13 ± 38.52); therefore it should be permissible to make the two materials into one. This has been done and the normal value then becomes 299.77 ± 18.72 with dispersion 193.66. As the figures show, dispersion round the normal value is great. There may be a great variation in the thrombin effect in different individuals, as also in the same individual, even on the same day. Thus, in exceptional cases, there may be values as low as 60 seconds and as high as 600 seconds.

The method error was calculated on 15 healthy men, aged about 30 years, with 20 double determinations. The first determination was made on blood from the right arm, and the second on blood from the left. The method error is ± 13.23 seconds or, in per cent of the normal value, 4.4 %.

As it may be difficult always to obtain uniform plasma, an investigation was made into whether there is a difference between different plasmas. The material consisted of 26 *partus cases*: on this were used plasmas with a sedimentation reaction of between 50 and 110 mm per hour (high s. r.), and those with sedimentation reaction of less than 10 mm per hour (low s. r.). The figures are given in table 17.

Table 17

Thrombin effect determined with different plasmas

$M \pm \varepsilon(M)$ = arithmetical mean ± standard error of the mean.
σ = standard deviation.

Number		$M \pm \varepsilon(M)$	σ
26	High s. r.	118.8 ± 18.1	92.5
26	Low s. r.	145.6 ± 19.3	98.5

The difference between high s. r. and low s. r. is 26.8 ± 26.5, implying no significant difference. Consequently, different plasmas can be used for testing purposes, without any influence on the value of the thrombin effect.

According to the table above, there is no significant difference between women and men. Studies into the influence of age reveal no significant differences. A difference is revealed by a comparison between the value calculated on 16 men aged 69—93 years, which value is 272.2 ± 19.4 with a dispersion of 77.5, and the normal value for 30-year-old men which is 287.47 ± 23.10, with a dispersion of 200.07; this difference is 15.27 ± 30.17 seconds, and consequently not significant.

To discover the behaviour of the thrombin effect at different times of the day and on different days, an investigation was made on 15 men of about 30 years of age, with samples taken at 7.30 a. m. (empty stomach) 12 noon and 6 p. m. on 3 days, with a day between each. The figures are given in table 18.

The difference between the morning and the noon value is 147.34 ±

Table 18

Thrombin effect at different times of the day

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

	Number	$M \pm \varepsilon(M)$	σ
7.30 a. m.	15	243.33 $\pm$ 38.92	150.72
12 noon	15	390.67 $\pm$ 48.44	187.59
6 p. m.	15	316.67 $\pm$ 72.18	279.57

62.13, and between the noon and the 6 p. m. value it is 74.00 $\pm$ 86.93; there is thus no significant difference in either case. Nor do the values for the different days show any significant differences. Nevertheless, though no such differences exist, it may be pointed out that there is a difference between the fasting value of the morning and that at 12 noon which is more than twice the standard error.

As the method of defibrination may be thought to influence the thrombin effect, a comparison has been made by defibrinating the same blood both by hand, by whisking with a glass rod, and with the defibrinator described earlier on. The material for this calculation consisted of 16 men over 69 years of age. The values are given in table 19.

Table 19

Thrombin effect determined on blood defibrinated by hand and with apparatus

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

	Number	$M \pm \varepsilon(M)$	σ
By hand	16	234.41 $\pm$ 16.8	67.0
With apparatus	16	272.20 $\pm$ 19.4	77.5

The deviation of 37.8 $\pm$ 25.7 thus shows there to be no difference between these values. For practical reasons, therefore, the defibrination has always been done with the apparatus.

A comparison between different test substances shows that none more suitable than citrated plasma is to be obtained. Experiments have been made with fibrinogen solution (produced according to Thordarson) + buffer, as also with ordinary citrated plasma +

buffer, but without yielding any improvement. The thrombin effect of the same blood has been investigated with these test substances in a material consisting of 15 partus cases. See table 20.

Table 20

Thrombin effect determined with different test substances

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

	Number	$M \pm \varepsilon(M)$	σ
Citrated plasma	15	128 $\pm$ 15.8	61.4
Citrated plasma + buffer	15	155 $\pm$ 19.5	75.4
Fibrinogen + buffer	15	180.3 $\pm$ 17.5	67.8

As citrated plasma gives the most distinct clots, this substance has always been used for testing purposes. The fibrinogen solution ought not to be used, as there is obviously difficulty in obtaining the same concentration. Two fibrinogen solutions produced according to the same method on two different occasions gave considerably divergent values when compared.

The influence of different temperatures has also been studied. A sample of 12 cm³ blood was taken; of this 5 cm³ was defibrinated, centrifuged, kept and examined at 22°, and 5 cm³ was similarly treated at 37°. The result is given in table 21.

Table 21

Thrombin effect determined at different temperatures

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

	Number	$M \pm \varepsilon(M)$	σ
Temp. 22°	45	243.33 $\pm$ 24.15	162.03
Temp. 37°	45	518.78 $\pm$ 46.82	314.06

The difference of 275.45 $\pm$ 52.68 is significant. Both the standard error and the dispersion are greater at 37°, so that determinations are always made at 22°; another reason for so doing is that the temperature in a hospital usually keeps round about this number of degrees. Should the room temperature be lower than this, a water bath at 22° is used in examining and storing.

CHAPTER IV

Adhesion Value and Number of Mast Cells

Operation and Adhesion Value

As appears from the survey of the literature, the adhesion tendency in leukocytes has been investigated only in a small number of cases. It has been claimed that the adhesion value is not influenced by narcosis, but is influenced by the operation. It furthermore seems likely that the adhesion tendency reaches a maximum value 3—4 days after the operation. A possible relation between this rise and thrombosis has been proposed, since the latter complication usually appears at the time in question.

As the histological structure of the thrombus shows that leukocytes are to be found along the thrombocyte beams, it seems reasonable to assume a connection between a raised adhesion tendency and thrombosis. This reason and the small amount of attention paid to the subject has prompted the investigation of the conditions in point on a more extensive material.

The adhesion value has been determined on a total of 163 persons, before and after operation. A survey of the results is given in tables 22 and 23.[1]

For the sake of further clarity, a graph is given in fig. 7.

In this material, the difference to be found between the preoperative values of men and women is not significant, being 0.30 ± 0.21. This does not, of course, rule out the possibility of obtaining a small significant difference on a larger material. In this connection, it should not be forgotten that we are not dealing here with a normal material, but with affected subjects. In other words, disease processes may possibly have affected the nature of the leukocytes. The material is not homogeneous as regards age or the character and extent of the

[1] Figures calculated from the first 107 cases have been used for these tables.

Table 22

Adhesion values in connection with operation

$M \pm \varepsilon(M)$ = arithmetical mean ± standard error of the mean.
σ = standard deviation.

Adhesion value	Men			Women		
	Number	$M \pm \varepsilon(M)$	σ	Number	$M \pm \varepsilon(M)$	σ
Before operation	49	6.99 ± 0.15	1.04	58	7.29 ± 0.15	1.15
Immediately after op.	49	6.47 ± 0.20	1.38	55	6.13 ± 0.23	1.74
2 hrs after op.	49	6.31 ± 0.22	1.55	55	6.17 ± 0.24	1.80
4 » » »	49	6.37 ± 0.22	1.54	55	6.10 ± 0.20	1.47
6 » » »	49	6.44 ± 0.23	1.58	55	6.15 ± 0.18	1.33

Table 23

Adhesion values in connection with operation

$M \pm \varepsilon(M)$ = arithmetical mean ± standard error of the mean.
σ = standard deviation.

Adhesion value	Morning value			Evening value		
	Number	$M \pm \varepsilon(M)$	σ	Number	$M \pm \varepsilon(M)$	σ
M e n						
1st day after op.	49	6.80 ± 0.20	1.42	47	6.83 ± 0.21	1.43
2nd » » »	49	7.03 ± 0.18	1.25	49	7.06 ± 0.19	1.35
3rd » » »	49	7.12 ± 0.18	1.25	47	7.05 ± 0.18	1.23
4th » » »	47	7.06 ± 0.13	0.92	46	7.15 ± 0.15	1.04
5th » » »	45	7.13 ± 0.18	1.22	43	7.18 ± 0.17	1.10
6th » » »	42	7.25 ± 0.17	1.10	37	7.15 ± 0.20	1.22
7th » » »	37	7.06 ± 0.21	1.26	32	6.94 ± 0.25	1.40
8th » » »	25	7.19 ± 0.29	1.46	15	6.67 ± 0.10	0.37
9th » » »	13	7.12 ± 0.35	1.25	—	—	—
W o m e n						
1st day after op.	54	6.46 ± 0.17	1.27	53	6.58 ± 0.20	1.43
2nd » » »	55	6.58 ± 0.19	1.40	52	6.81 ± 0.22	1.59
3rd » » »	52	6.86 ± 0.17	1.26	52	6.88 ± 0.18	1.31
4th » » »	52	6.82 ± 0.16	1.18	50	6.83 ± 0.17	1.18
5th » » »	51	7.05 ± 0.18	1.25	50	7.13 ± 0.13	0.93
6th » » »	48	7.00 ± 0.15	1.02	39	6.84 ± 0.19	1.20
7th » » »	40	7.06 ± 0.19	1.22	26	7.25 ± 0.23	1.18
8th » » »	25	7.08 ± 0.20	0.99	23	7.17 ± 0.23	1.09
9th » » »	22	7.07 ± 0.20	0.93	18	7.17 ± 0.26	1.09
10th » » »	17	7.14 ± 0.26	1.08	12	7.10 ± 0.31	1.07

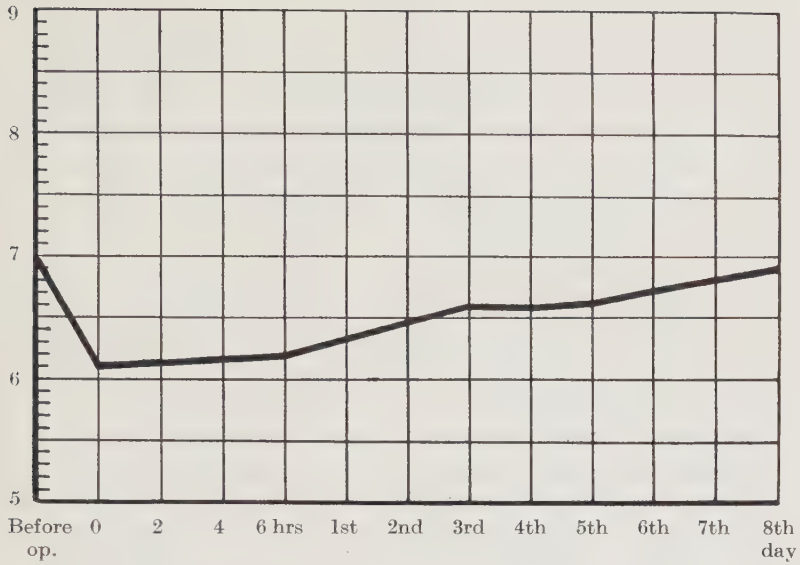

Fig. 7. Adhesion values in connection with operation.

surgical measures. It is therefore by no means surprising that certain differences can be seen in the preoperative values, and that the normal value which the author was able to discover in 50 conscripts shows a significant difference. The difference between this normal value and the preoperative value is for men 1.73 ± 0.16. This emphasizes the fact that the operation material cannot be considered as representative of normal material. It is impossible to say definitively whether the fact that conscripts are younger than the average population played any very great part.

As regards the postoperative values, we can in the first place establish that the standard deviation rises. This increase is significant for both men and women at all the junctures investigated, i. e. up to 6 hours after the operation. It can also be seen that women showed significant differences between pre- and postoperative values up to 2 days after the operation, after which time the values seemed to fall slowly to normal. The figures for men follow the same course. However, the differences are not significant for the men, but then, nor are there any significant differences between men and women. The different conditions among the sexes has partly to do with the fact that the material for women is somewhat larger; the difference may well also be due in part to pure factors of chance, above all to the

fact that the preoperative value for men is lower than that for women. In view of the general conformity of the course followed by the values. however, it should be possible to say with certainty that, both in men and in women, the operation is immediately followed by lowered values, which persist for some hours, but which then gradually return to normal within a week; the material is not large enough to determine precisely when this takes place.

The figures in table 23 give the adhesion value calculated both morning and evening at about the same times, from day to day; an inspection of these figures is unable to reveal any difference between the two values. The slight differences which occasionally occur are in no case significant.

The material has been analysed by a division into major and minor operations; the results are given in tables 24 and 25.

A first inspection of these tables shows that the initial values for the two groups are not quite the same. The difference is not significant. however, and may well be due to chance. Both the values are above the normal; this, as has already been suggested, is because the material consists of affected subjects. The course of the figures is interesting. In both cases the values fall in the hours immediately after the opera-

Table 24

Adhesion values in connection with minor operations

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Adhesion value	Number	$M \pm \varepsilon(M)$	σ
Before operation	34	7.01 ± 0.29	1.70
Immediately after op.	34	6.76 ± 0.28	1.64
2 hrs » »	34	6.68 ± 0.35	2.02
4 » » »	34	6.56 ± 0.30	1.77
6 » » »	34	6.46 ± 0.32	1.87
1st day » »	34	6.55 ± 0.31	1.82
2nd » » »	32	7.03 ± 0.29	1.65
3rd » » »	32	6.75 ± 0.33	1.89
4th » » »	32	6.76 ± 0.30	1.68
5th » » »	28	7.09 ± 0.28	1.47
6th » » »	26	6.92 ± 0.38	1.92
7th » » »	22	6.77 ± 0.23	1.81

Table 25

Adhesion values in connection with major operations (not infected)

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Adhesion value	Number	$M \pm \varepsilon(M)$	σ
Before operation	112	6.13 ± 0.14	1.49
0 hrs after »	113	5.07 ± 0.17	1.76
2 » » »	113	4.95 ± 0.16	1.70
4 » » »	112	4.98 ± 0.16	1.67
6 » » »	109	5.01 ± 0.16	1.72
1st day after op.	110	5.34 ± 0.17	1.75
2nd » » »	109	5.54 ± 0.17	1.79
3rd » » »	109	5.73 ± 0.18	1.87
4th » » »	108	5.86 ± 0.15	1.56
5th » » »	103	6.00 ± 0.17	1.75
6th » » »	95	6.00 ± 0.16	1.60
7th » » »	72	6.04 ± 0.19	1.60
8th » » »	48	6.38 ± 0.22	1.55
9th » » »	35	6.28 ± 0.29	1.74

tion. Only in the group Major operations, which is somewhat larger, are the differences statistically significant; they are also more marked here, as would be expected. It is of course not possible to determine, without further investigation, the reason for the difference. The narcosis might be thought to play a part, but earlier investigations tend to show this is not so. It therefore seems most natural to assume that the actual operation is the cause, i. e. that the difference is contingent on tissue lesions, loss of blood, and the like. If this is so, a major operation would obviously have a more distinct effect than a minor one. (It might be mentioned that the material under Major operations comprises the following types: stomach resections, uterus amputations, salpingoophorectomies, cholecystectomies, nephrectomies, and a number of resections on intestines and stomach for cancer.)

The group in the above material has been divided into age groups, and their median calculated. The pre- and postoperative adhesion value was then calculated in the same way as was previously used for the two age groups. The results of these calculations are given in table 26.

Table 26

Adhesion values and age

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Adhesion value	< 44.8 years			>44.8 years		
	Number	$M \pm \varepsilon(M)$	σ	Number	$M \pm \varepsilon(M)$	σ
Before operation	51	5.72 ± 0.20	1.41	48	6.48 ± 0.19	1.32
0 hrs after »	52	4.49 ± 0.22	1.60	48	5.40 ± 0.25	1.75
2 » » »	52	4.32 ± 0.20	1.42	48	5.37 ± 0.22	1.50
4 » » »	52	4.42 ± 0.19	1.38	48	5.39 ± 0.22	1.56
6 » » »	50	4.35 ± 0.22	1.56	46	5.59 ± 0.23	1.55
1st day after op.	52	4.67 ± 0.19	1.35	46	5.96 ± 0.23	1.54
2nd » » »	52	4.92 ± 0.21	1.53	45	5.98 ± 0.28	1.89
3rd » » »	49	5.08 ± 0.24	1.67	47	6.10 ± 0.29	1.96
4th » » »	50	5.33 ± 0.20	1.43	45	6.00 ± 0.26	1.71
5th » » »	46	5.50 ± 0.24	1.61	44	6.26 ± 0.30	1.97
6th » » »	42	5.47 ± 0.24	1.52	38	6.42 ± 0.28	1.74
7th » » »	35	5.64 ± 0.27	1.57	35	6.06 ± 0.30	1.76
8th » » »	20	5.64 ± 0.32	1.42	29	6.62 ± 0.30	1.64
9th » » »	13	5.34 ± 0.56	2.01	23	6.34 ± 0.31	1.50
10th » » »	9	(5.78)		17	6.48 ± 0.34	1.40

The preoperative values will be found to differ only slightly from one another. A comparison with the normal value (5.6 ± 0.05) shows the latter to be somewhat lower. The difference is 0.12 ± 0.21 for younger subjects, and 0.88 ± 0.20 for older subjects. No difference is to be found between the two groups, and they present the same course as those previously illustrated in fig. 7—i. e. a fall after the operation, with a gradual return to normal. Both groups yield significant differences between post- and preoperative values.

There is little evidence of a connection between adhesion values and thrombosis. The age group which according to the literature is most subject to thrombo-embolism, i. e. the older one, shows adhesion values at operation which deviate least from the normal. However, several operation cases complicated with thrombosis have been submitted to a closer study in respect of the adhesion value. The figures are given in table 27.

Not even these figures indicate any connection with thrombosis. It is, however, plain that the adhesion value deviates in certain cases

Table 27

Adhesion values in thrombo-embolism cases (1—8). The black figures show when the thrombo-embolism was diagnosed

Adhesion value		1	2	3	4	5	6	7	8
Before operation		6.7	7.4	6.7	5.3	7.1	8.3	7.3	6.8
Immediately after op.		6.1	5.4	6.2	5.0	6.9	8.2	7.4	5.7
2 hrs	» »	5.8	5.4	5.7	6.8	7.5	8.3	7.3	5.7
4 »	» »	5.0	6.4	4.9	5.1	8.2	8.7	7.0	6.1
6 »	» »	5.1	6.2	5.2	5.6	9.0	8.3	7.2	5.7
1st day after op.	P. M.	6.6	6.2	6.6	5.3	8.8	9.0	8.0	5.7
	A. M.	6.6	**5.0**		5.4	8.8		7.2	6.0
2nd » » »	P. M.	7.0	5.4	6.1	5.5	8.5	**9.0**	6.9	5.0
	A. M.	**6.6**	5.6		5.2	8.9		6.2	5.5
3rd » » »	P. M.	7.3	5.0	**8.2**	6.3	9.0		6.0	6.9
	A. M.	7.6	5.0		6.1	6.2		6.0	5.4
4th » » »	P. M.	6.7	4.6	9.2	7.3	8.1		5.7	**6.6**
	A. M.	6.9	5.3		7.1	7.9		6.4	6.3
5th » » »	P. M.	7.0	5.2	6.1	8.2	7.3		6.5	6.9
	A. M.	7.0	5.2		8.1	7.1		**6.3**	6.6
6th » » »	P. M.	7.2	6.5		**7.6**	**7.4**		6.4	7.2
	A, M.	6.9	6.1		7.7	7.3		6.2	6.7
7th » » »	P. M.	7.2	6.4		7.9	7.5		6.9	7.1
	A. M.	7.3	6.3		7.3			5.7	7.2

from the normal, whereas in others it is not affected at all. Needless to say, a connection is conceivable, but a large material would be needed to prove this. A reasonable conjecture is that changes in the adhesion value in connection with thrombosis are present only for very short periods, and that they were consequently unable to manifest themselves in these few cases. The fact remains that the determination of the adhesion values in thrombosis has not yielded any practicable aide to the diagnosis of the individual case.

Operation and Number of Mast Cells

It has been shown by Swedish authors that heparin is bound to the mast cells of tissue. Furthermore, a few cases of thrombosis of the retinal vessels have been found where the mast cells along the

walls of the thrombotic vessels were, abnormally, lacking in heparin. An explanation offered (Holmgren) is that the production of heparin had for some reason been checked, and that this then caused thrombosis. It has also been found that single cases of myeloic leukaemia involves an increase of the blood's mast cell value, prolonged coagulation time being then demonstrable.

One would be very inclined to link up a lowered heparin production with thrombosis. If there is a connection between the two, it might reasonably be assumed that, when the conjunction is in force, the tissue's and also the blood's mast cells (which seem to be related) decrease in number or completely disappear. Although the hypothesis lacks adequate foundation, the effect of an operation on the blood mast cells has been investigated, as this might well be presumed to help on an assessment of the problem.

Table 28 gives figures from 37 cases of major operations often followed by thrombosis.

Table 28

Number of mast cells in % by operations

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Number of mast-cells in %	Morning value			Evening value		
	Number	$M \pm \varepsilon(M)$	σ	Number	$M \pm \varepsilon(M)$	σ
Before operation	37	0.378 ± 0.054	0.33	—	—	—
Immediately after op.	37	0.243 ± 0.039	0.23	—	—	—
2 hrs » »	37	0.173 ± 0.029	0.18	—	—	—
4 » » »	37	0.182 ± 0.034	0.20	—	—	—
6 » » »	37	0.200 ± 0.031	0.18	—	—	—
1st day after op.	37	0.173 ± 0.036	0.22	36	0.127 ± 0.025	0.15
2nd » » »	37	0.295 ± 0.057	0.34	35	0.224 ± 0.037	0.22
3rd » » »	35	0.190 ± 0.041	0.24	35	0.185 ± 0.044	0.26
4th » » »	35	0.275 ± 0.059	0.35	31	0.282 ± 0.056	0.31
5th » » »	31	0.316 ± 0.056	0.31	31	0.247 ± 0.048	0.27
6th » » »	31	0.322 ± 0.058	0.32	29	0.378 ± 0.058	0.31
7th » » »	29	0.318 ± 0.043	0.23	27	0.317 ± 0.035	0.18
8th » » »	19	0.257 ± 0.035	0.15	21	0.356 ± 0.055	0.25
9th » » »	10	0.200 ± 0.033	0.10	10	0.416 ± 0.063	0.20
10th » » »	8	(0.260)	.	8	(0.438)	.

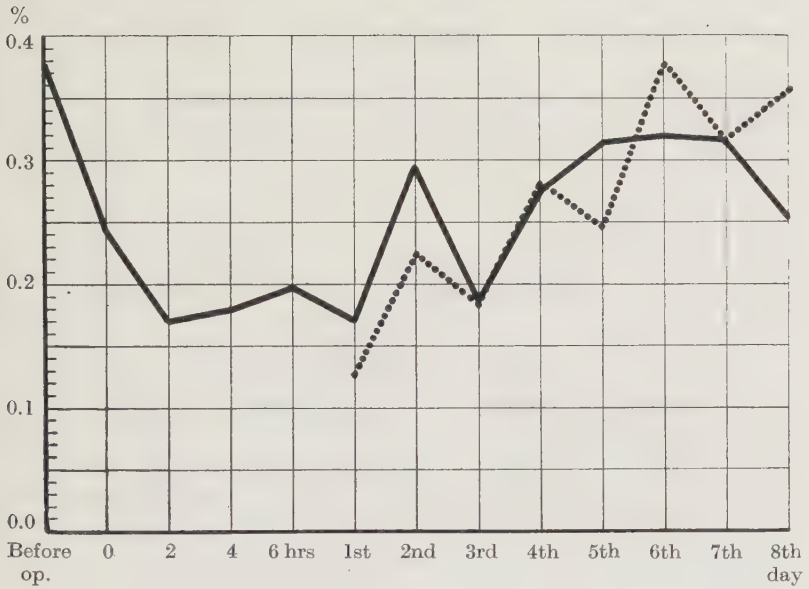

Fig. 8. Number of mast cells in % by operation.
Whole line = morning value; dotted line = evening value

When looking at the figures, the preoperative value seems to be higher than normal (0.181 ± 0.019 %), and the difference which is 0.197 ± 0.057 is therefore significant.

The dispersion in the operation material is considerably greater than in the normal material, no doubt indicating greater inhomogeneity. No reason for this can be found, but it might be mentioned that the normal value has been calculated on young healthy subjects, whereas the value in question here concerns affected subjects of all ages. The graph in fig. 8 shows a considerable drop from the preoperative value as early as 2 hours after the operation. The difference is 0.205 ± 0.06, and therefore significant. From this juncture the value slowly rises, to have passed the normal and reached the preoperative one on the 5rd—6th day. In view of the large method error, individual cases can hardly be assessed by single determinations of the mast cell value. No further investigations have therefore been made.

Coagulation Time of the Blood

Coagulation Time and Operation

As has been mentioned earlier on, the changes in coagulation time which are attendant on operations and partus have been studied by a number of investigators, but their results do not always agree, so that the question cannot be considered settled. The acceptable assumption of a connection between increased coagulation tendency (decreased coagulation time) and thrombosis has also been discussed.

Table 29

Time of coagulation in connection with operations

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Time of coagulation	Men			Women		
	Number	$M \pm \varepsilon(M)$	σ	Number	$M \pm \Sigma(M)$	σ
Before operation	16	389.7 ± 6.7	26	71	393.8 ± 2.2	18
Immediately after op.	22	356.4 ± 9.0	42	88	367.2 ± 2.7	25
2 hrs » »	60	348.1 ± 5.2	40	108	350.5 ± 2.9	34
4 » » »	53	312.7 ± 5.7	42	102	321.5 ± 5.2	53
6 » » »	65	317.9 ± 5.4	44	108	341.5 ± 4.8	55
1st day after op.	58	383.6 ± 3.9	30	108	384.0 ± 2.0	22
2nd » » »	55	393.5 ± 2.9	22	107	391.0 ± 1.8	19
3rd » » »	53	392.8 ± 3.0	22	108	389.1 ± 2.0	21
4th » » »	56	393.8 ± 3.1	23	104	392.8 ± 1.7	17
5th » » »	56	396.6 ± 2.4	18	106	391.7 ± 1.7	17
6th » » »	52	392.3 ± 3.2	24	85	391.6 ± 1.9	17
7th » » »	58	395.3 ± 2.4	19	87	391.7 ± 2.2	20
8th » » »	52	393.8 ± 3.1	22	79	389.9 ± 2.2	19
9th » » »	46	395.5 ± 2.6	17	76	392.7 ± 2.2	19
10th » » »	35	393.1 ± 3.9	23	59	396.8 ± 2.4	18

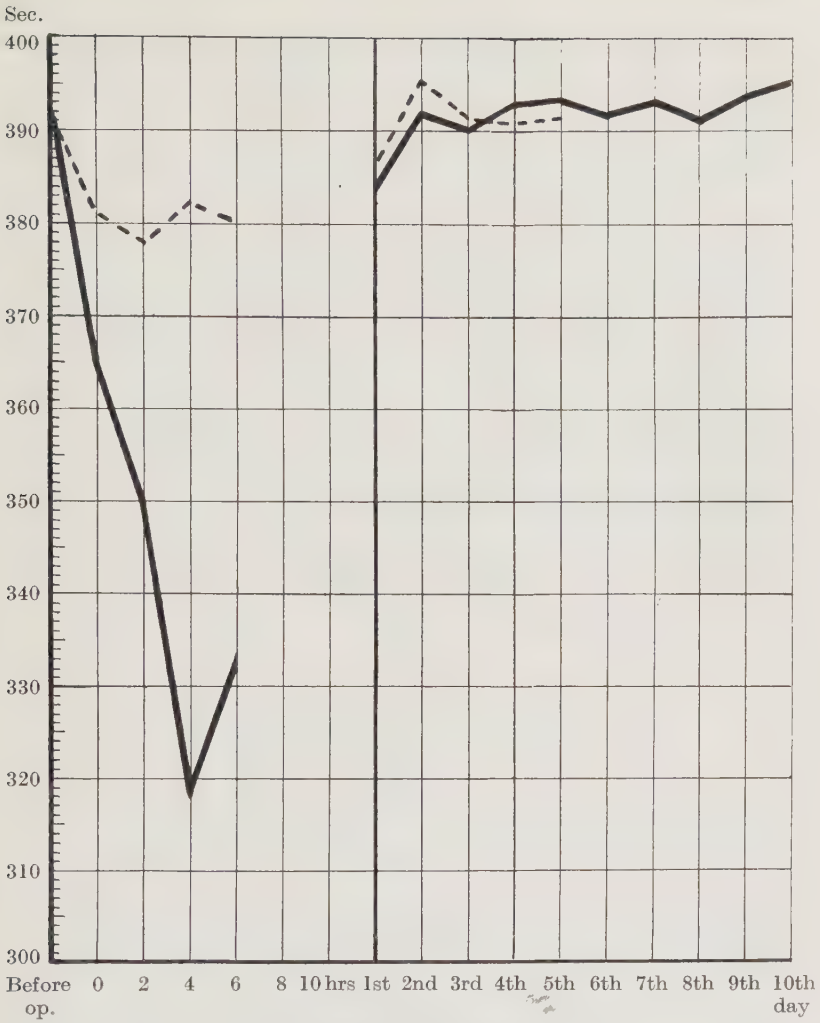

Fig. 9. Time of coagulation.
Whole line = in connection with operation.
Dotted line = » » » partus.

but this problem, too, has not been fully clarified by publications to date. An attempt has been made to get to grips with the above questions by the collection and analysis of a material described more fully in Chap. II. This material was first divided up into men and women; the figures for the calculations made are given in table 29. A graph in fig. 9 illustrates the figures more clearly. The table

and the graph show that there is a slight fall in the coagulation time, i. e. an increased coagulation tendency in the blood, after operations.

However, this change in coagulation can be observed only in the hours immediately following the operation; the very next day the values are fairly normal. A closer inspection of the figures shows the coagulation tendency to be already stronger immediately after the operation; the difference between the values *before* and immediately *after* the operation is 33.3 ± 11.2 seconds for men, and 26.6 ± 3.5 seconds for women. This difference is further accentuated during the next few hours, and the lowest value for the coagulation time appears 4 hours after the operation. After this juncture the value gradually rises to normal. A comparison of the figures for men and women yields no significant difference. Nor are any such differences to be found between men and women in the other determinations. It therefore seems permissible to make the materials for men and women into one, even though a small difference between the two

Table 30

Time of coagulation and age

$M \pm \varepsilon(M)$ = arithmetical mean ± standard error of the mean.
σ = standard deviation.

Time of coagulation	< 45 years			> 45 years		
	Number	$M \pm \varepsilon(M)$	σ	Number	$M \pm \varepsilon(M)$	σ
Before operation	28	396.6 ± 3.8	20	18	392.2 ± 6.5	27
Immediately after op.	23	349.6 ± 7.0	33	23	360.7 ± 6.2	30
2 hrs » »	43	338.8 ± 5.4	35	57	341.2 ± 5.1	38
4 » » »	43	324.0 ± 5.8	32	57	312.3 ± 4.9	37
6 » » »	47	329.9 ± 7.5	51	63	325.8 ± 6.8	54
1st day » »	44	391.1 ± 3.3	22	63	385.9 ± 2.8	22
2nd » » »	42	399.2 ± 2.2	15	55	389.6 ± 2.8	21
3rd » » »	43	393.6 ± 3.2	21	55	389.0 ± 3.3	24
4th » » »	42	396.5 ± 2.8	18	60	392.8 ± 2.9	23
5th » » »	41	397.0 ± 2.8	18	56	394.3 ± 2.5	18
6th » » »	39	395.3 ± 3.2	20	49	391.3 ± 2.9	20
7th » » »	41	392.0 ± 2.9	19	53	390.9 ± 3.7	27
8th » » »	34	395.7 ± 2.3	13	51	389.8 ± 4.1	29
9th » » »	32	399.1 ± 2.8	16	48	393.2 ± 2.9	19
10th » » »	28	397.0 ± 2.8	15	42	393.1 ± 4.2	27

may not be altogether out of the question, and this has been done. in order to give a smaller standard error. The values for the coagulation time have then been plotted on a graph. An inspection of the figures for determinations from the 1st to the 10th day inclusive shows there to be no significant average differences of the coagulation time.

Some of the material comprising only major stomach operations has been divided into 2 groups, according to age, with the dividing-line at 45 years. The figures in table 30 for calculations according to this division shows there to be no difference between the 2 groups. Individuals on either side of the dividing-line react alike with a fall in the coagulation time after operations. There are significant differences between pre- and postoperative values, as soon as the operation is over. The values fall steadily during the next few hours, reaching a minimum after 4 hours. The 6th hour after the operation already sees an initial return to normal, however, which is reached on the following day. The investigation made here has thus been unable to show any difference for the coagulation tendency in older and younger individuals.

Coagulation Time and Delivery

Table 31 gives the figures for determinations of the coagulation time in partus cases. The figures are also plotted graphically in fig. 9.

A comparison between these figures and those in table 29 shows that the coagulation time is clearly not so affected by partus as by operation. A graph illustrating this is found in fig. 9. The lowest value is found 2 hours after partus; the difference between this value and that before delivery is 14.3 $\pm$ 4.8 seconds, i. e. practically significant. The period after partus shows, otherwise, no significant differences from the value before partus. If the value for the coagulation time before an operation is compared with the same value before partus, they are found to agree well.

Thus, it may be definitely asserted that the coagulation time is affected by the operation, but this does not establish whether it is the tissue lesion, the narcosis or the loss of blood that is the main

Table 31

Time of coagulation in connection with partus

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Time of coagulation	Number	$M \pm \varepsilon(M)$	σ
Before delivery	21	391.4 ± 3.7	17
Immediately after d.	27	381.1 ± 3.1	16
2 hrs after d.	27	378.0 ± 3.1	16
4 » » »	27	382.2 ± 2.7	14
6 » » »	21	380.2 ± 3.8	17
1st day after d.	26	386.1 ± 3.4	17
2nd » » »	20	395.2 ± 3.5	16
3rd » » »	25	391.4 ± 3.1	15
4th » » »	20	390.8 ± 3.7	17
5th » » »	22	391.4 ± 2.7	13

cause. It has nevertheless been maintained in the literature that certain facts point to a considerable influence from both the narcosis and the loss of blood. This idea is the more acceptable in that the coagulation tendency reaches more or less normal values no longer than 8 hours after the operation, i. e. the juncture when, in most cases, the narcosis and the effects of loss of blood (Sahli's experiment) have worn off. This being so, and provided that thrombosis is contingent on the coagulation time of the blood, it should not be wrong to suppose that the thrombi are formed during the time now under discussion: after all, it covers two of the known factors considered to influence thrombosis, i. e. blood change (first and foremost increased coagulation tendency) and retardation of the stream in the veins (cf. Frimann-Dahl). However, the blood stream in the veins is retarded even more some days after the operation, i. e. during the period when the coagulation tendency is normal. Furthermore, thromboses usually manifest themselves during these days. We might expect the coagulation tendency of the blood in *thrombosis cases* to increase afresh in the days following the operation, or else to find that it was greater than normal from the very beginning, i. e. before the operation, and that these circumstances induce thrombosis when added to the slow blood stream. One possibility is that the thrombosis always begins during the period immediately following the operation

Table 32

Time of coagulation in thrombo-embolism cases

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

No.	The day before thrombosis	The day of thrombosis	The day after thrombosis
1	370	360	360
2	370	330	240
3	240	200	240
4	150	130	—
5	130	120	130
6	255	255	255
7	315	300	270
8	350	300	315
9	370	375	370
10	375	370	—
11	315	320	315
12	300	240	360
13	300	150	255
14	290	180	210
15	285	240	240
	4415	3870	3560
$M \pm \varepsilon(M)$	294 $\pm$ 19	258 $\pm$ 22	274 $\pm$ 19
$\sigma =$	76	87	69

and, if this condition persists longer than normal or if increased coagulation again sets in, the thrombi are so built upon that they occlude the vascular lumen and give a clinically manifest thrombosis.

Table 32 gives the figures for 15 thrombosis cases, the compilation being made in respect of the time thrombosis (embolism) set in. Only the day before and the day after thrombosis has been given here, as the material is not sufficient for a statistical calculation of more than these days. An examination of the figures shows a considerable fall in the coagulation time, as compared with the normal value, during all 3 days. If, however, the individual cases are examined, figures are found which are only slightly less than the normal value. Plainly, then, the thrombosis in a number of cases is allied with an increased coagulation tendency in the blood. However, the figures hardly appear to be such as aid the diagnosis of thrombosis from determina-

tions of the coagulation time: high, practically normal values may be found, despite the presence of thrombosis. This may be because the coagulation time is subject to rapid changes; if the determination is made at a time when the coagulation is slow, a lower value need not appear, though we would expect it to do so as a rule in a fairly large material.

CHAPTER VI

Thrombin Effect

Thrombin Effect and Vitamin C Standard

The investigation of the normal variation in the thrombin effect has revealed great individual differences (cf. Chap. III). Even before this, however, a high value had on one occasion been obtained during the treatment of a pneumonia case. Subsequent determinations for this case continued to give high values. Prompted by this, more detailed investigations were made to clarify the situation. The cause of the relatively strong thrombin effect might have been thought to lie in 1) the disease as such, 2) a complication with thrombosis, 3) treatment with sulfathiazol or 4) treatment with Vitamin C. In theory, of course, a combination of these factors, or some of them, might plausibly be supposed responsible.

An investigation of a number of other pneumonia cases yielded no rise in the thrombin effect. It was therefore considered *a priori* improbable that the disease as such could have caused the rise. There were no clinical signs of thrombosis in the case with raised thrombin effect. It therefore seemed most natural to assume that the effect was linked up with the treatment, and the first steps taken were consequently to test the separate effects of sulfathiazol and Vitamin C.

Sulfathiazol and Vitamin C were given separately to 5 experiment subjects, the thrombin effect being studied at the same time. The sulfathiazol did not seem to influence this effect in these subjects, whereas there was an appreciably enhanced effect in those given surplus Vitamin C (10 cgm thrice daily for 5 days). After these preliminary tests, the effect of Vitamin C was tested on 20 healthy subjects aged 20—30 years: 12 men and 8 women. They all received 10 cgm of Vitamin C in tablet form 3 times daily, and the urine was investigated every day with the dichlorphenolindophenol test. The experi-

ment was made during the month of April, when the Vitamin C standard can be assumed to be low. On the 5th day 14 of the test subjects showed saturation, i. e. they began to excrete Vitamin C with the urine. The remaining 6 did not show saturation till the 8th day. When a subject was saturated, the thrombin effect was determined (the test substance used was plasma from the same person, who had not received extra Vitamin C). 50 cgm of Vitamin C were injected intravenously in one dose, and the thrombin effect determined anew 15 minutes later. Finally, a determination was made 5 days later.

The results of this experiment are given in table 33, and illustrated by a graph in fig. 10.

Table 33

Thrombin effect and Vitamin C

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Thrombin effect	Number	$M \pm \varepsilon(M)$	σ
Before Vitamin C	20	250.5 $\pm$ 40.2	180
Saturated	20	90.8 $\pm$ 10.2	46
$^1/_4$ hour after 50 cgm Vitamin C	20	64.0 $\pm$ 9.4	42
5 days later	16	185.9 $\pm$ 32.4	130

A survey of the figures shows that the value before the beginning of the experiment was 250.5 $\pm$ 40.2, i. e. somewhat higher than normal. There is no significant difference, however, and it is probable that the slight deviation is due to chance. The dispersion is considerable, being 180. After saturation there is a thrombin effect which is as high as 90.8 seconds, and exhibits fairly slight dispersion in the series. Intravenous injection of 50 cgm of Vitamin C is followed by a further increase in the effect after 15 minutes, though the difference from the saturation value is not significant. After this juncture the effect falls, but has still not reached the pre-experimental value by the 5th day. It is thus clear that Vitamin C increases the thrombin effect.

The effect of Vitamin C in coagulation systems used here may, however, be linked up either with the thrombin or with the fibrinogen.

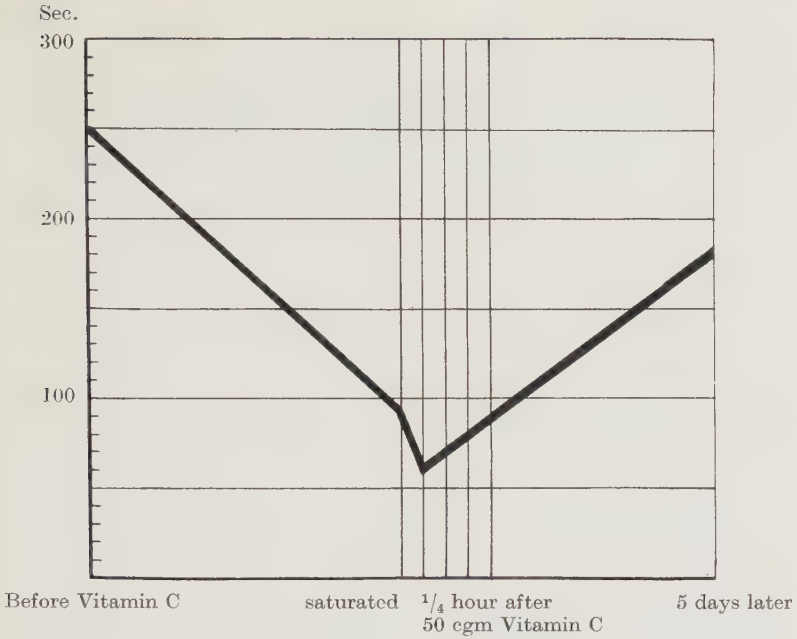

Fig. 10. Thrombin effect and Vitamin C.

Heat coagulation of the fibrinogen was therefore investigated in citrated plasma, as complementary test. 0.2 cm³ was added to 0.2 cm³ physiological salt solution. Further, 0.2 cm³ plasma was added to 0.2 cm³ of a Vitamin C solution (5 cgm Na-salt of the ascorbic acid. dissolved in 1.2 ml. $p_H = 5.5$). Finally, 0.2 cm³ plasma was added to 0.2 cm³ of a stronger Vitamin C solution (50 cgm Na-salt of the ascorbic acid dissolved in 5 ml. $p_H = 5.5$). The coagulation was induced by lowering the test-tubes into a water bath at 55° C. 20 tests were made. The results are given in table 34.

The figures show that Vitamin C retards the heat coagulation of the fibrinogen, and that increased concentration of Vitamin C prolongs the coagulation time.

Needless to say, this experiment does not warrant the definite conclusion that the Vitamin C *in vivo* has an inhibiting effect on the fibrinogen's coagulation, though it seems most natural to assume that such is the case. However, it has been previously shown by experiment (Marx and Bayerle) that, when the same amount of a thrombin solution of known strength is given to plasma taken from animals suffering from scurvy and to plasma from other animals with a

Table 34

Heat coagulation of fibrinogen in citrated plasma and in citrated plasma + Vitamin C solution of different strength

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Time of coagulation	Number	$M \pm \varepsilon(M)$	σ
0.2 cm³ citrated plasma + 0.2 cm³ physiol. salt sol.	20	90.0 ± 4.6	20
0.2 cm³ citrated plasma + 0.2 cm³ Vitamin C sol. 5 cgm/1.2 ml.	20	118.6 ± 6.7	30
0.2 cm³ citrated plasma + 0.2 cm³ Vitamin C sol. 50 cgm/5 ml.	20	155.4 ± 9.3	41

normal Vitamin C standard, the plasma of the former coagulates more quickly than that of the latter. These experiments therefore support the view that the Vitamin C standard has an effect on the transformation of fibrinogen.

If the fibrinogen in the plasma coagulates more quickly when there is Vitamin C deficiency (coagulation of the fibrinogen in the plasma of scorbutic animals by standard thrombin solution), but normal plasma on the other hand coagulates with particular rapidity on the administration of serum (thrombin effect) which has been acted upon by Vitamin C, this must obviously be because Vitamin C in some way potentizes the thrombin effect, this then outweighing the inhibitory action that Vitamin C has on the coagulatory powers of the plasma fibrinogen. It might also be mentioned that the amount of Vitamin C which proved to affect the thrombin in the studies on the thrombin effect, and which can be assumed to affect the fibrinogen (0.2 cm³ of vitiminized serum added to 0.2 cm³ of citrated plasma), is very slight compared with the amount of Vitamin C in the above *in vitro* tests.

A further series of experiments has been made. In these the thrombin effect was determined in 16 persons, using for test substance both normal plasma and plasma acted upon by Vitamin C. Both types of plasma were taken from the same person. First, the normal plasma was prepared (1 part 3.7 % citrate solution and 4 parts blood),

whereupon 1 gm Vitamin C was given intravenously. 10 minutes later new citrated plasma was made in the same way. The figures are high throughout (= lower thrombin effect) for the tests where Vitamin C plasma was used, but there is no significant difference between them and those where normal plasma was used. Thrombin effect determined with normal plasma gives the mean 280 $\pm$ 16, with a dispersion of 64; determination with plasma acted upon by Vitamin C gives the mean 300 $\pm$ 19, with a dispersion of 75. Difference between the two values = 20 $\pm$ 24 and therefore not significant.

The tests which were made should warrant the conclusion that the thrombin effect is raised by the administration of Vitamin C, and further that, on heat coagulation of fibrinogen, the addition of Vitamin C in the concentration used here has an inhibitory effect.

The experiments made here suggest that a high content of Vitamin C in the blood should make for special liability to thrombosis, provided that high thrombin effect induces or is a necessary condition for thrombosis. However, this circumstance can hardly be of any very great importance for the occurrence of thrombosis under ordinary conditions, as Vitamin C surplus of the extent in question here is not normally frequent, and is certainly not found in diseased states.

Thrombin Effect and Delivery

As it seems reasonable to assume a certain relation between strong thrombin effect and thrombosis, and as thrombosis is a relatively common occurrence during the period immediately following pregnancy, it is obviously of interest to try and get an idea of the thrombin effect in association with partus. 82 newly delivered women have therefore been investigated in some detail for changes in the thrombin effect. For technical reasons (injuries to the veins) samples have been taken only before partus and immediately after its conclusion, with further samples 6 hours after delivery, and subsequently once daily until the patient got up. That is to say, no investigations were made during the 6 hours immediately following partus. The first sample was usually taken 12 hours before delivery, at earliest. As the patients were in hospital, this means that labour had already begun. The results of this investigation are given in table 35, and graphically illustrated in fig. 11.

Table 35

Thrombin effect in connection with partus

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Thrombin effect	Number	$M \pm \varepsilon(M)$	σ
Before delivery	52	141.1 ± 8.7	62
Immediately after d.	35	130.4 ± 15.0	89
6 hrs after d.	43	144.1 ± 11.7	76
1st day after d.	75	127.0 ± 8.6	75
2nd » » »	77	127.3 ± 8.4	74
3rd » » »	77	148.4 ± 9.0	78
4th » » »	78	130.1 ± 8.7	77
5th » » »	76	118.8 ± 8.7	76
6th » » »	74	110.1 ± 6.7	57
7th » » »	60	144.2 ± 12.1	93
8th » » »	34	130.7 ± 9.4	55

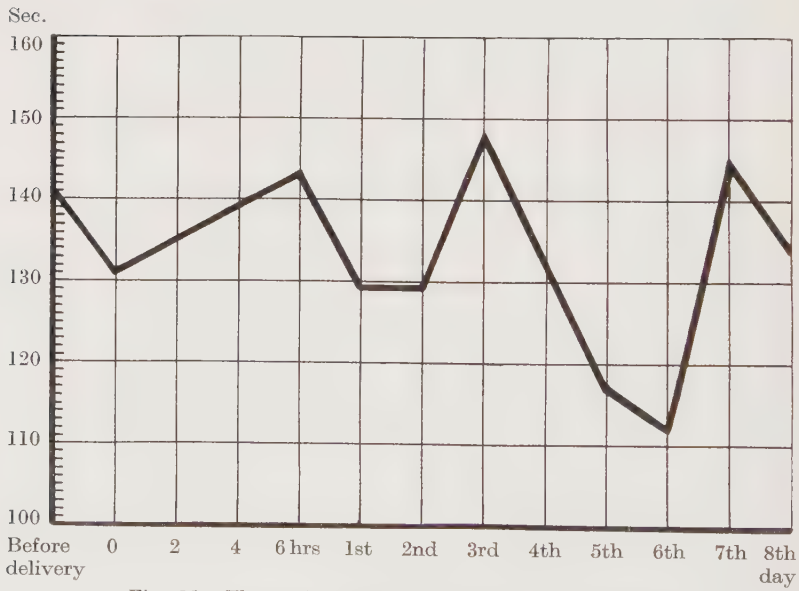

Fig. 11. Thrombin effect in connection with partus.

The figures in the table show that the value of the thrombin effect before partus is considerably higher than the normal value (299.8 ± 18.7). The difference of 158.7 ± 20.6 is significant. The probable

reason for this is that, according to what has been shown earlier on (Thordarson), the prothrombin content reaches its maximum value during the time mentioned above. The first few days after partus present considerable deviations from the value before partus; however, since the normal material also shows a considerable dispersion, the fluctuations which the curve (see fig.) presents here may well be due in part to chance. It is not possible to determine whether the peculiar peaks of the curve indicate a special causal connection; a larger material would be necessary to establish this. On the 6th day after partus, however, there is an appreciable rise in the value, i. e. greatly increased thrombin effect. The difference between the value before partus and that on the above-mentioned day is 31.0 ± 11.0—that is to say, to all intents and purposes significant. It might be said of the thrombin effect that it is stronger than normal even before partus, and that it subsequently shows an increased effect with a maximum value on the 6th day after delivery.

The material has here been divided up into 2 groups, one containing primiparae and the other multiparae. The figures are given in table 36.

Table 36

Thrombin effect in partus cases. The material has been divided into primiparae and multiparae

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Thrombin effect	Primiparae			Multiparae		
	Number	$M \pm \varepsilon(M)$	σ	Number	$M \pm \varepsilon(M)$	σ
Before delivery	27	123.2 ± 13.0	67	25	160.6 ± 10.6	53
Immediately after d.	13	120.8 ± 20.8	75	22	136.1 ± 20.8	97
6 hrs » »	16	143.1 ± 19.8	79	25	149.8 ± 15.2	76
1st day » »	37	123.8 ± 13.6	83	38	132.1 ± 11.1	69
2nd » » »	38	125.3 ± 12.9	80	39	129.4 ± 12.1	76
3rd » » »	38	133.9 ± 12.2	75	39	166.0 ± 13.7	86
4th » » »	37	120.5 ± 11.2	68	41	139.0 ± 13.6	85
5th » » »	39	118.1 ± 12.9	80	37	121.4 ± 11.1	71
6th » » »	38	103.4 ± 9.2	57	36	119.6 ± 10.2	61
7th » » »	36	136.5 ± 9.6	57	24	155.8 ± 27.0	132
8th » » »	21	125.0 ± 12.9	59	13	140.0 ± 13.3	48

A survey of the table shows that the figures for multiparae are throughout higher than those for primiparae, though in no case are the differences significant.

Thus, the investigation has shown that pregnancy at the beginning of partus (early labour) is associated with increased thrombin effect, and also that partus acts on the thrombin effect by raising it still further, this rise being most pronounced on the 6th day after delivery. The above assumption of a certain relation between the thrombin effect and thrombosis hereby receives support, since thrombosis has already been shown to set in as a rule at this juncture, i. e. 6 days after partus. Before this question is discussed in detail, however, we will touch on the thrombin effect and its changes in association with operation.

Operation and Thrombin Effect

It has already been shown, both in the survey of the literature and in the chapters of this thesis which describe blood investigations carried out, that an operation is associated with blood changes; it was therefore considered of interest to study the thrombin effect in surgical intervention and see whether this effect, too, shows deviations from the normal value. A material consisting of 67 operated cases (47 men and 20 women) has been studied in some detail to this end. As constant punctures injure the veins unduly, it has as a rule been necessary to restrict the determinations to before the operation, immediately after it, 6 hours after it, and then each subsequent day until the patient got up. This he usually did about 7 days after the operation. The pre-operative value was determined on the same day as the operation was performed, generally an hour or so before it began.

A calculation of the material divided according to sex was first made. The figures are outlined in table 37.

An inspection of the table shows that the pre-operative values are higher than normal (299.8 ± 18.7), but only the women show the difference (81 ± 25) from the normal to be significant. This may be partly because the female material is considerably smaller than the male. The strong thrombin effect before the operation is, however, no doubt mainly due to the fact that affected individuals are in question

Table 37

Thrombin effect in connection with operation. The material has been divided into men and women

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Thrombin effect	Men			Women		
	Number	$M \pm \varepsilon(M)$	σ	Number	$M \pm \varepsilon(M)$	σ
Before operation	47	271.9 ± 20.1	138	20	218.7 ± 16.0	72
Immediately after op.	31	288.4 ± 20.5	114	15	252.7 ± 20.2	78
2 hrs after op.	11	287.6 ± 37.0	123	1	(340)	.
4 » » »	3	(229.3)	.	1	(440)	.
6 » » »	19	274.4 ± 26.8	117	5	(315)	.
1st day » »	46	259.9 ± 19.5	132	22	238.6 ± 23.0	108
2nd » » »	42	242.9 ± 16.4	106	19	192.5 ± 27.3	119
3rd » » »	43	233.1 ± 19.9	130	20	200.8 ± 22.5	101
4th » » »	35	187.5 ± 12.1	72	18	200.8 ± 20.0	85
5th » » »	37	217.2 ± 16.4	100	15	224.7 ± 35.1	136
6th » » »	37	212.4 ± 18.7	114	17	198.6 ± 13.5	55
7th » » »	29	227.5 ± 17.2	93	11	202.2 ± 29.6	99
8th » » »	10	205.0 ± 28.5	90	4	(181.3)	.
9th » » »	7	(211.4)	.	3	(243.3)	.
10th » » »	7	(190.7)	.	2	(257.5)	.

here. Nevertheless, the course of the figures agrees fairly well in the two series, and no juncture covered by the investigation shows significant differences; it should therefore be permissible to make the two materials into one. Needless to say, minor differences between men and women might be expected when calculating a larger material, but they can hardly be of any very marked character. The men and women have therefore been made into one single material, to give a smaller standard error.

A calculation of the figures from this larger material gives approximately the same deviations as are to be traced in table 37. The results of this calculation are given in table 38, and the figures are illustrated by the graph in fig. 12.

If we look first at the pre-operative value, the thrombin effect seems to be stronger here than in normal material (299.8 ± 18.7). The difference, 44 ± 24, is not significant, however, and may possibly be due to chance. Nor do the deviations, present immediately after the

Table 38

Thrombin effect in connection with operation

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Thrombin effect	Men + women		
	Number	$M \pm \varepsilon(M)$	σ
Before operation	67	256.0 ± 15.0	123
Immediately after op.	46	276.7 ± 15.4	105
2 hours after op.	12	291.9 ± 34.1	118
4 » » »	4	(282.0)	.
6 » » »	24	283.0 ± 22.0	108
1st day after op.	68	253.0 ± 15.0	123
2nd » » »	61	227.2 ± 14.2	110
3rd » » »	63	222.8 ± 15.2	121
4th » » »	53	192.0 ± 10.3	75
5th » » »	52	219.4 ± 15.1	109
6th » » »	54	208.0 ± 13.3	98
7th » » »	40	220.5 ± 14.8	94
8th » » »	14	198.2 ± 24.2	91
9th » » »	10	221.0 ± 24.1	76
10th » » »	9	(205.6)	.

operation and lasting till 6 hours later, show significant differences from the pre-operative value; they, too, are probably due to chance. The following days see successive deviations, and on the 4th day the deviation becomes significant, i. e. 64.0 ± 18. The thrombin effect can be taken to behave as follows: before the operation and during the time immediately following— approximately up to and including the first day after — it does not change at all, or only very slightly. After this juncture it begins to increase, and is strongest on the 4th day after the operation. After this time it gradually decreases and approaches the normal value. The material is not sufficiently large for us to decide when this value is reached.

A remarkable circumstance, showing a deviation from the type of other blood changes associated with operations, is that the thrombin effect does not seem to come under any immediate influence from the operation trauma, the lesion to the tissues, and the narcosis. During the time when the other blood changes studied deviate most greatly from the pre-operative value — i. e. the first 5 —6 hours after opera-

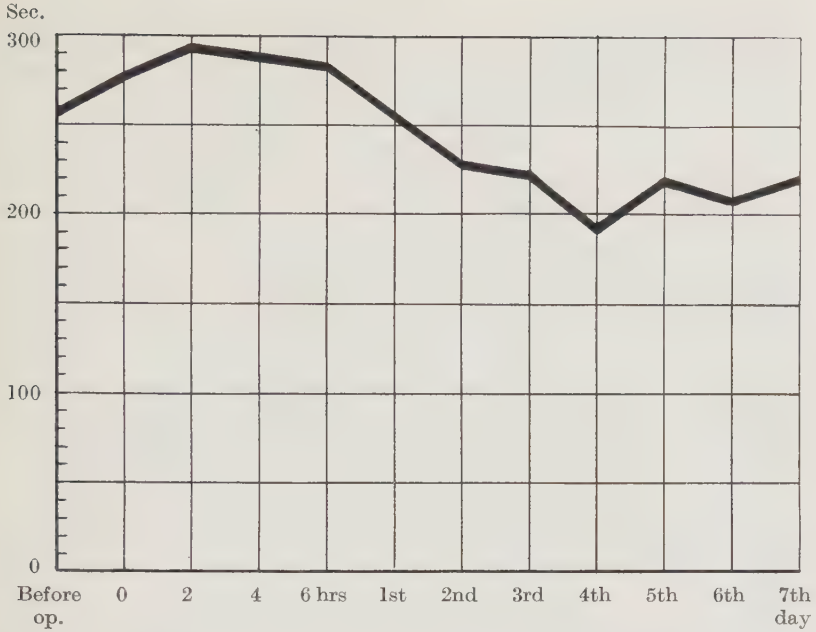

Fig. 12. Thrombin effect in connection with operation.

tion—the thrombin effect is hardly influenced. The earlier finding that the coagulation time showed the greatest deviation immediately after operation had led one to expect parallel changes for the thrombin effect. It is not easy to explain the actual difference between these two findings, dealing as they do with intimately connected blood conditions. It should, however, be borne in mind that, in the determination of the coagulation time, thrombin begins to form as soon as the blood has left the organism, and that the thrombin has its effect on the fibrinogen within 5–6 minutes, after which time the coagulation is usually completed. Here, the time factor depends on the rate at which the thrombin is formed, whereas this does not have any influence on the value when determining the thrombin effect. When this latter method of investigation is used, the thrombin probably takes the same time to form as in the determination of the coagulation time; on the other hand, 30 minutes elapse before it exerts its effect on the fibrinogen. On this account, a factor (antithrombin) is here active in the process, which factor cannot have an influence in the coagulation of the blood. The antithrombin, that is to say, can only take full effect on the thrombin after a certain incubation period, which amounts to at least 15 minutes.

In the light of these facts, the above-mentioned discrepancy in the figures for the coagulation time and the thrombin effect are explicable. The former's great deviation immediately after the operation can hardly be due to anything else than abnormally rapid formation of thrombin—i. e. increased amount of thrombokinase, as the thrombin effect is not affected at this juncture. This increased quantity of thrombokinase must derive from the lesion caused by the operation to the tissue. The dissimilarity occurring at a later point after the operation, with strong thrombin effect and at the same time fairly normal coagulation time, is no doubt due to the fact that the thrombin forms at a more or less normal rate, but the inhibiting antithrombin effect is weak. The stronger thrombin effect may, of course, be attributed to an increased quantity of thrombin. If this were so, however, it would probably also have an enhancing effect on the coagulation tendency of the blood. But this is not so, so that this assumption does not seem here to cover the actual state of affairs.

However, an increased quantity of prothrombin, and therewith an increased amount of thrombin, may also be expected to occur without any noteworthy curtailment of the coagulation time for whole blood. If the amount of thrombokinase is small, the coagulation of the fibrinogen in the whole blood can probably be completed before all the prothrombin has been converted into thrombin. The thrombin increase is hereby concealed in the determination of the coagulation time, but appears in the determination of the thrombin effect. We have therefore to take into account two factors which may give a stronger thrombin effect —namely decreased antithrombin effect and increased amount of thrombin, or a combination of both.

Thrombosis and Thrombin Effect

The literature shows that the theories on the mechanism of thrombosis have been dominated by different factors during different periods of time. For a long time the mechanical factor was taken to be the deciding one, and the conditions of the blood stream, slow stream, or different types of stream induced by obstructions in its path or by injuries to the intima, were considered to be the main

causes of thrombosis. In more recent literature attempts have been made to show that changes in the composition of the blood also affect the formation of thrombi, and several authors have been inclined to stress these changes.

It must be presumed that, if the operation is sufficiently extensive, it is most often followed by a certain circulatory insufficiency. A natural result of this is that the blood flows more slowly in the vessels, above all in the veins of the legs, for the mere recumbency occasions here a lower rate of stream than normally (Frimann-Dahl). However, the slow stream can by itself hardly be the cause of thrombosis: it is sometimes seen how, after operation, the failing circulation leads to death, without thrombosis being present. It may further be mentioned that traumatic shock may sometimes be accompanied by total stagnation of the blood in certain vascular regions, and yet thrombi are not formed on that account. On the other hand, it is clear that a slow blood stream should promote thrombosis more than a rapid one. The slow stream means that the different constituents of the thrombus are more abundantly present in the blood's peripheral section –that is to say, beside the vascular wall where the thrombus is first formed. Different vortices may also be expected to have the same effect. Furthermore, an incipient thrombus can be washed away by a rapid stream. If the thrombus begins to form on account of a local impulse proceeding from the vascular wall, then a slow blood stream should also favour thrombosis, by the fact that the effect per blood unit then becomes lengthier.

However, blood changes cannot be assumed to be the sole causes of thrombosis, either. This idea is untenable because, if such were the case, it would mean a total thrombosis of the entire mass of blood. It can, however, be assumed that blood changes should have a favourable effect on the formation of thrombi, and that when local changes are present, a thrombus is formed. Favourable conditions of this kind are local retardation of the stream and possibly a change in the type of the stream, the formation of vortices and the like, or a lesion to the intima with the giving off of thrombokinase.

The aim of the present work has been to obtain a more differentiated picture of these blood changes, nevertheless taking into account that they cannot be of crucial importance but must be combined with local factors. According to what has been shown previously, increased coagulation tendency in the blood seems to bear a relation to the

thrombus, although normal values may be present in individual cases. However, the juncture when the coagulation tendency is greatest after operation is that covering the first 4—5 hours — that is to say, a juncture when thrombo-embolisms appear only by way of exception. As has already been indicated, a deposition of the thrombus may naturally take place at this juncture, with successive deposits, and manifestation on a subsequent occasion. Nevertheless, coagulation, and probably the formation of thrombi also, are relatively rapid processes, so that, in the vast majority of cases, it is hardly likely that thrombi are formed in the manner sketched here. A more reasonable assumption seems to be that the thrombus is formed some days later in the after-course of the operation. As has been shown, a blood change having its maximum value during this time is the thrombin effect. But before a thrombus (coagulation) can arise, another factor must contribute, namely thrombokinase. When the endothelium is injured, thrombokinase may be formed which can act on the prothrombin present in the blood flowing past. It is also conceivable that the thrombokinase is formed from the corpuscular elements in the blood if retarded stream or vortices cause these to accumulate, and thus die off from exposition to inanition or, from some other cause, disintegrate and give off thrombokinase. Now, if the thrombin effect of the blood is strong, due either to high thrombin content or a low antithrombin effect or to a combination of both, thrombosis will be set up in the presence of the above-mentioned favourable factors. On the other hand, if the thrombin effect is weak or the stream rapid, or if the coagulation tendency has been lowered by heparin, the defences of the organism have time to work, so that even the thrombokinase, and at any rate the formed thrombin, is neutralised or excreted from the organism. Working on this line of thought, the thrombin effect has been more closely studied in a number of thrombosis cases. The aim was to make clear whether the changes in the thrombin effect bear a relation to thrombosis, and whether a determination of this contributes to a diagnosis. As has previously been stated, the value of the thrombin effect presents a considerable dispersion round the normal value. This, together with the further circumstance that the thrombin effect is influenced by Vitamin C, must be taken into account when determining individual values. It has been possible to investigate 17 cases of certain thrombosis in more detail. The thrombosis was

localized in the veins of the legs, and in all cases there was pronounced phlegmasia alba dolens. The thrombosis set in 6—10 days after operation or childbirth, as the case might be. Two cases in the Medical Dept. (11 and 17) had, before the onset of the thrombosis, shown values for the thrombin effect varying between 250—400.

An inspection of the figures in the table below clearly shows that thrombosis is combined with a strong thrombin effect, persisting for several days. On the day before thrombosis sets in the thrombin effect is already 96, and on the actual day it has risen to 51. It should be mentioned here that the diagnosis of thrombosis in these cases could not be made until the thrombus had obturated the vascular lumen. A thrombus of these dimensions could hardly form

Table 39

Thrombin effect in connection with thrombo-embolism

$M \pm \varepsilon(M)$ = arithmetical mean $\pm$ standard error of the mean.
σ = standard deviation.

Thrombo-embolism cases No.	The day before thrombosis	The day of thrombosis	1st day after thrombosis	2nd day after thrombosis	3rd day after thrombosis	4th day after thrombosis
1	170	30	58	45	80	90
2	30	25	40	50	55	80
3	110	30	60	85	—	—
4	105	50	20	190	50	105
5	30	30	30	35	30	35
6	34	34	47	95	50	32
7	30	50	60	70	125	245
8	210	60	65	200	135	155
9	60	70	50	50	45	75
10	60	30	30	60	70	205
11	100	60	30	40	70	125
12	55	60	—	135	—	—
13	90	90	85	100	—	—
14	210	30	50	190	210	—
15	105	35	80	45	70	50
16	60	50	50	110	130	160
17	165	130	85	90	80	110
$M \pm \varepsilon(M)$	96 $\pm$ 14.7	51 $\pm$ 6.6	53 $\pm$ 5.0	94 $\pm$ 13.4	86 $\pm$ 19.9	113 $\pm$ 17.9
σ	61	27	20	55	48	64

instantaneously or in the space of a few hours, so that the high values of the thrombin effect for the day before thrombosis was diagnosed must in several cases be assumed to have been determined at a juncture when the thrombosis had already set in, although it was not showing any current clinical symptoms. If the individual figures are inspected it is seen that those for the day before the manifestation of thrombosis show the limit values 210 and 30. The weakest thrombin effect (210) is thus above the normal value. The weakest thrombin effect (130) on the day thrombosis set in is a value often found in childbirth cases, and sometimes in operations. If the value on the day thrombosis set in is compared with the highest values shown by childbirth cases, certain days for the latter show as high values as were measured for thrombosis. However, the thrombosis cases show a high level up to the 4th day after the disease manifested itself (only isolated cases can be studied after this juncture) whereas a strongly raised thrombin effect seems otherwise to be of more transient character. It seems, therefore, to have been made clear that thrombosis is associated with a strong thrombin effect usually under 100 (a value of 130 was obtained in one case), but an equally high effect may occasionally be measured in other pathological conditions also, and a thrombin effect of corresponding strength may even sometimes appear in healthy individuals. The thrombin effect is activated by Vitamin C, though there is probably a certain state of equilibrium *in vivo*, if real coagulation can occur here, since Vitamin C seems to retard the coagulation of the fibrinogen. However, the rate of the blood stream in healthy individuals is so high that it can be presumed to prevent the formation of thrombi. In cases of childbirth and operation, on the other hand, the conditions of the blood stream favour the onset of thrombosis (strain on the circulatory system + recumbency). A transient strong thrombin effect does not, consequently, seem able to induce thrombosis, whereas a longer period with high thrombin effect may do so. However, if we look at Case 14, we see that on the day before thrombosis set in the thrombin effect was measured at 210, whereas on the following day, when the thrombus was found, there was a thrombin effect of 30. Similar values are found in Cases 1 and 7. Although only 3 cases are involved, it can therefore be said that, in certain cases, thrombosis may be combined with a high thrombin effect lasting for a relatively short time (less than 24 hours). In the light of these figures and of the above-

mentioned conditions, high thrombin effect seems admittedly to be of importance for the formation of thrombi, but is probably not the decisive factor. It may equally well be a question of effect as of cause. Some other factor beyond those advanced here, i. e. retardation of the stream and increased thrombin effect, seems necessary to cause thrombosis. This factor is probably of local character, e. g. lesion to the intima or change of the stream's type in conformity with the theory put forward above.

A strong thrombin effect persisting longer than 24 hours thus suggests the presence of thrombosis within the organism, but there is no proof that such is the case. No positive diagnostic data can therefore be reached by determining the thrombin effect; negative data, on the other hand, can be obtained, for weak thrombin effect should exclude thrombosis. Values for the thrombin effect of less than 160 (i. e. numerically high values), above all if they show a falling tendency on a later occasion, rule out this disease. Thus, in doubtful cases, a determination of the thrombin effect helps to indicate the therapeutic treatment.

Summary

It transpires from the relatively full discussion of the literature on the subject that the onset of thrombosis is materially influenced by mechanical factors in the form of a generally retarded blood stream, together with local obstructions to its flow. Different investigations have been made to show that changes in the composition of the blood also have an influence, but it has not been possible to obtain any more definitive information. Nevertheless, it is known that the risk of thrombosis is considerably increased after operation and childbirth, and that these events are accompanied by blood changes concerning both the corpuscular elements and the plasma constituents. Two of the results of these changes are a somewhat raised sedimentation reaction and some increase of the coagulation tendency of the blood. However, this latter change can hardly be considered to have been definitively clarified by previous investigations. In the present work, the author has attempted to make a more all-round investigation of the composition of the blood in different respects after operation and childbirth.

1) The author has been able to show that the adhesion value of the leukocytes falls immediately after operation. The difference from the preoperative value 2 hours after the operation is 1.12 ± 0.28. This difference is gradually levelled out, and is gone after about 1 week. As was expected, major operations give larger differences in the adhesion value than minor ones. It may be mentioned that operation material showed no difference between morning and evening values; this is interesting because a number of authors have claimed to find a difference following the 24-hour rhythm in normal material. Furthermore, according to the material investigated by the author, age does not seem to exert any noteworthy influence.

2) The number of mast cells falls immediately after operation. The difference from the preoperative value 2 hours after operation is 0.205 ± 0.06, but the number begins to rise as little as 3—4 hours after, and preoperative values are again reached on the 5th—6th day.

3) The coagulation time of the blood decreases immediately after operation; this has been seen by earlier authors. This phenomenon apparently occurs even before the operation is over, as determinations made at the end show a significant difference, amounting to 33.3 ± 11.2 for men, and 26.6 ± 3.5 for women. However, the lowest value is found 4 hours after the operation, when the difference from the preoperative value is 77.0 ± 8.8 for men, and 72.3 ± 6.1 for women. The coagulation time soon reaches normal values, however, probably about the 2nd day. Here, too, the course of the figures does not seem to be at all influenced by age.

The coagulation time is also lowered in childbirth, though not so much as in operation. Here, the lowest value is reached as early as 2 hours after delivery. The difference from the preoperative value is 14.3 ± 4.8. Normal values are reached after 6—7 hours.

4) The thrombin effect is fairly unaffected in the time immediately following operation, but increases subsequently, to show a maximum value on the 4th day, with a difference from the preoperative value of 64 ± 18. The thrombin effect is stronger than normal during pregnancy, and increases still more after childbirth. It is strongest on the 4th—6th day. The increase of the thrombin effect after partus seems to be rather more pronounced in primiparae than in multiparae.

To study factors influencing the thrombin effect, investigations have also been made on persons both with low Vitamin C standard and after they had been saturated with Vitamin C; a difference could be seen here. The thrombin effect is far stronger in the saturated subjects, and the difference amounts to 159.7 ± 41.5 in the author's material. It has furthermore been found that the heat coagulation of the fibrinogen *in vitro* is slower in the presence of increased quantities of Vitamin C.

5) When investigating cases of thrombosis, the author has not found any very marked difference, suitable for diagnostic purposes, in the adhesion value of the leukocytes. As regards the coagulation time, it has been found to be shorter than normal in certain cases of thrombosis, but this is not a regular occurrence, so that this blood change has no diagnostic value, either; moreover, it is hardly likely that this curtailment is of crucial importance for the onset of thrombosis. On the other hand, all the patients with thrombosis (17 cases in all) have shown a strongly increased thrombin effect, which might

amount to 6 times the normal strength, though there are a few exceptions, with not so markedly high values. This strong thrombin effect may well be of diagnostic value in as much as it usually appears before clinical symptoms of thrombosis manifest themselves. From a diagnostic viewpoint, therefore, it should be interesting to study the thrombin effect at an earlier stage. If it is not raised, this would argue against thrombosis. On the other hand, there may be strong thrombin effect without thrombosis necessarily setting in. (High values may in exceptional cases be seen in association with operation and childbirth in subjects apparently free from thrombosis, and occasionally in healthy individuals.)

It is to a certain extent surprising that the thrombin effect may be increased postoperatively, although the coagulation time shows no corresponding increase. In Chap. VI the author discusses those possibilities of interpreting this phenomenon which most readily present themselves. But in any case, the situation seen here should be of clinical interest, independent of the theoretical interpretation.

Finally, it should be stated that the observed changes in the adhesion tendency of the leukocytes and the length of the coagulation time seem not to be so important as regards the time at which the thrombus is formed. The raised thrombin effect coincides better as to time with the juncture when thrombosis sets in. On the other hand, it is perhaps not impossible that the raised effect is a secondary occurrence in the formation of the thrombus.

Literature

Adler, L.: Arch. f. Gynäk. 95: 349, 1911.

Allen, E. V.: Arch. Surg. 15: 254, 1927.

Aschoff, L.: Virchows Arch. f. path. Anat. 130: 93, 1892.

» Beitr. z. path. Anat. u. z. allg. Path. 52: 205, 1912.

» Deutsche med. Wchnschr. 38: 2057, 1912.

» Ztschr. f. Kreislaufforsch. 26: 363, 1934.

Aschoff, L., Beck, B. v., de la Camp, O., & Krönig, B.: Beiträge zur Thrombosefrage. Leipzig 1912.

Astrup, P.: Heparinundersögelser. Diss. Köbenhavn 1944.

Astrup, T., & Darling, S., Acta physiol. Scandinav. 3: 168, 1942.

Barta, I., & Jakab, M.: Deutsches Arch. f. klin. Med. 164: 350, 1929.

Bauer, G.: Svenska läk.-tidn. 40: 2849, 1943.

» Acta chir. Scandinav. 90: 229, 1944.

Bauer, G., & Jorpes, E.: Nord. med. 22: 1003, 1944.

Baumgarten, P.: Zentralbl. f. d. med. Wissensch. 15: 131, 1877.

Bayer, F. W., & Philipsborn, E. v.: Folia haemat. 60: 69, 1938.

Beneke, R.: Handbuch der allgemeinen Pathologie. 2: 2: 130. Leipzig 1913.

Bergquist, G.: Acta chir. Scandinav. 83: 415, 1940.

Best, C. H.: Brit. M. J. 2: 977, 1938.

Best, C. H., Cowan, C., & MacLean, L. D.: Science 85: 338, 1937.

Bizzozero, J.: Virchows Arch. f. path. Anat. 90: 261, 1882.

Borgström, S.: Acta chir. Scandinav. 89: 68, 1943.

Boshamer, K.: Klin. Wchnschr. 6: 740, 1927.

Brinkhous, K. M., Smith, H. P., & Warner, E. D.: Am. J. M. Sc. 193: 475, 1937.

Brinkhous, K. M., Smith, H. P., Warner, E. D., & Seegers, W. H.: Am. J. Physiol. 125: 683, 1939.

Bültemann, H.: Zentralbl. f. Gynäk. 53: 1218, 1929.

Butt, H. R.: J. A. M. A. 113: 390, 1939.

Butt, H. R., Snell, A. M., & Osterberg, A. E.: J. A. M. A. 113: 383, 1939.

Cannon, W. B., & Gray, H.: Am. J. Physiol. 34: 232, 1914.

Chargaff, E., Ziff, M., & Moore, D. H.: J. Biol. Chem. 139: 383, 1941.

Charles, A. F., & Scott, D. A.: J. Biol. Chem. 102: 425 and 431, 1933.

» Biochem. J. 30: 1927, 1936.

Chauvin, E., Esmenard, & Jaur: Gynéc. et obst. 13: 123, 1926.

Crafford, C.: Acta chir. Scandinav. 79: 407, 1937.

» *ibid.* 82: 319, 1939.

Crafford, C., Nord. med. 2: 1231, 1939.
» *ibid.* 2: 1040, 1939.
» Acta med. Scandinav. 107: 116, 1941.
» Nord. med. 13: 285, 1942.
» *ibid.* 16: 2836, 1942.
Culp, O. S.: Bull. Johns Hopkins Hosp. 67: 1, 1940.
Dahlberg, G.: Statistical Methods for Medical and Biological Students. London 1940.
Dahl-Iversen, E., & Ramberg, E.: Hospitalstid. 75: 371, 1932.
de Courcy, J. L.: Anesth. & Analg. 8: 342, 1929.
Denecke, K.: München. med. Wchnschr. 76: 1912, 1929.
Dienst, A.: Arch. f. Gynäk. 65: 369, 1902.
» *ibid.* 86: 314, 1908.
» Ztschr. f. Geburtsh. u. Gynäk. 82: 103, 1920.
Dietrich, A.: München. med. Wchnschr. 67: 931, 1920.
» *ibid.* 76: 272, 1929.
Eagle, H.: J. Gen. Physiol. 18: 531, 547 & 813, 1935.
Eberth, C. J., & Schimmelbusch, C.: Die Thrombose nach Versuchen und Leichenbefunden. Stuttgart 1888.
Eufinger, H.: Arch. f. Gynäk. 149: 414, 1932.
Eufinger, H., & Kiltz, W.: Monatschr. f. Geburtsh. u. Gynäk. 99: 279, 1935.
Evans, W. H.: Proc. Roy. Soc. Med. 22: 729, 1929.
Fahr, Th.: Klin. Wchnschr. 6: 2179, 1927.
Fåhraeus, R.: Biochem. Ztschr. 89: 355, 1918.
» Acta med. Scandinav. 55: 1, 1921.
» Klin. Wchnschr. 7: 100, 1928.
Fleig, C., & Lefébure, M.: J. de physiol. et de path. gén. 4: 615, 1902.
Foà, C.: Zentralbl. f. Physiol. 27: 603, 1913.
Fonio, A., & Schwendener, J.: Die Thrombocyten des menschlichen Blutes. Bern 1942.
Frimann-Dahl, J.: Acta chir. Scandinav. 76: Suppl. 36, 1935.
Frykholm, R.: Nord. med. 4: 3534, 1939.
Fuld, E., & Spiro, K.: Beitr. z. chem. Phys. u. Path. 5: 171, 1904.
Garrot-Allen, J., & Livingstone, H.: Arch. Surg. 42: 522, 1941.
Gasser, H. S.: Am. J. Physiol. 42: 378, 1917.
Geissendörfer, H.: Deutsche Ztschr. f. Chir. 246: 88, 1935.
Geissendörfer, R.: Thrombose und Embolie. Leipzig 1935.
Goosens, H., & Liebich, H. A.: Arch. f. klin. Chir. 195: 106, 1939.
Hammarsten, O.: Ztschr. f. physiol. Chem. 22: 333, 1896.
» *ibid.* 28: 98, 1899.
Hankin, E. H.: Zentralbl. f. Bakt. 12: 777 och 809, 1892.
Hanssen, P.: Acta chir. Scandinav. 80: 340, 1938.
Hare, T., & Glynn, E. E.: Proc. Roy. Soc. Med. 22: 741, 1929.
Hartmann, E.: Klin. Wchnschr. 11: 1828, 1932.
Hartmann, J.: München. med. Wchnschr. 56: 796, 1909.
Hedenius, P.: Über wahre Metachromasie der weissen Blutkörperchen. Diss. Stockholm 1940.

Hellsten, W. O.: Phlebographic Studies and Heparin Treatment in Thrombo-Embolic Diseases. Diss. Stockholm 1942.
Hermanson, Å.: Svenska läk.-tidn. 22: 27, 1925.
Heusser, H.: Deutsche Ztschr. f. Chir. 210: 132, 1928.
Holmgren, H.: Ztschr. f. wissensch. Mikr. 55: 419, 1938.
Holmgren, H., & Stenbeck, A.: Acta ophth. 18: 271, 1940.
Holmgren, H., & Wilander, O.: Ztschr. f. mikr.-anat. Forsch. 42: 242, 1937.
Holmgren, H., & Wohlfart, G.: Nord. med. 11: 2771, 1941.
Höring, O.: Deutsche Ztschr. f. Chir. 207: 360, 1928.
Horwitz, S.: Ztschr. f. d. ges. exper. Med. 57: 380, 1927.
Howell, W. H.: Am. J. Physiol. 29: 187, 1911—12.
Howell, W. H., & Holt, E.: Am. J. Physiol. 47: 328, 1918.
Hueck, H.: Arch. f. klin. Chir. 136: 774, 1925.
Jaschke, R. Th. v.: Arch. f. Gynäk. 129: 951, 1926.
Jorpes, E.: Heparin. Its Chemistry, Physiology and Application in Medicine. London 1939.
Jorpes, E., Holmgren, H., & Wilander, O.: Ztschr. f. mikr.-anat. Forsch. 42: 279, 1937.
Jürgens, R.: Deutsche med. Wchnschr. 65: 124, 1939.
Kaae, S.: Nord. med. 20: 2215, 1943.
Kaneko, S.: Arch. f. klin. Chir., 197: 395, 1939.
Keller, R.: Arch. f. Gynäk. 97: 540, 1912.
Klemensiewicz, R.: Beitr. z. path. Anat. u. z. allg. Path. 63: 321, 1916.
Klotz, L.: Klin. Wchnschr. 11: 1536, 1932.
König, W.: Klin. Wchnschr. 9: 2060, 1930.
Kristenson, A.: Acta med. Scandinav. 69: 453, 1928.
» Upsala läkaref. förh. 43: 169, 1938.
Krukenberg, H.: Monatschr. f. Geburtsh. u. Gynäk. 85: 374, 1930.
Kugelmass, I. N.: Arch. internat. de physiol. 21: 139, 1923.
Küster, H.: Ergebn. d. inn. Med. u. Kinderh. 12: 666, 1913.
Kuwaschima, K.: J. Biochem. 3: 91, 1923.
Lampert, H.: Die physikalische Seite des Blutgerinnungsproblems. Leipzig 1931.
Landsberg, M.: Biochem. Ztschr. 50: 245, 1913.
Lehmann, J.: Nord. med. 12: 3192, 1941.
Leissner, S. H.: Nord. med. 13: 523, 1942.
Lenggenhager, K.: Deutsche Ztschr. f. Chir. 244: 77, 1934.
» Helvet. med. acta 1: 527, 1935.
» Mitt. a. d. Grenzgeb. d. Med. u. Chir. 44: 175, 1936.
» Über die Entstehung, Erkennung und Vermeidung der postoperativen Fernthrombose. Leipzig 1941.
Linde, S.: Acta chir. Scandinav. 84: 310, 1940.
Lindsay, E. C.: Lancet, 208: 327, 1925.
Lockhart-Mummery, J. P.: Proc. Roy. Soc. Med. 22: 740, 1929.
Löhr, W.: Deutsche Ztschr. f. Chir. 183: 1, 1923.
Lucadou, W. v.: Ztschr. f. Kreislaufforsch. 25: 785, 1933.
Manstein, B.: Deutsche med. Wchnschr. 69: 104, 1943.

Marx, R., & Bayerle, H.: Biochem. Ztschr. 315: 366, 1943.
Mason, M. F.: Surgery 5: 451 and 618, 1939.
Mc Clintock, J. T., & Magers, E.: Proc. Soc. Exper. Biol. & Med. 24: 203, 1926—27.
Mellanby, J.: J. Physiol. 38: 441, 1909.
Mendenhall, W. L.: Am. J. Physiol. 38: 33, 1915.
Morawitz, P.: Deutsches Arch. f. klin. Med. 79: 1, 1903.
» *ibid.* 79: 215 & 432, 1904.
» Beitr. z. chem. Phys. u. Path. 5: 133, 1904.
» Ergebn. d. Physiol. 4: 307, 1905.
Murray, D. W. G., Jaques, L. B., Perrett, T. S., & Best, C. H.: Canad. M. A. J. 35: 621, 1936.
» Surgery 2: 163, 1937.
Naegeli, O., Blutkrankheiten und Blutdiagnostik. 5. Aufl. Berlin 1931.
Neu, M., & Kreis, P.: München. med. Wchnschr. 58: 2441, 1911.
Nicolaysen, E.: Acta chir. Scandinav. 69: 21, 1931.
Nielsen, P. H.: Nord. med. 16: 2882, 1942.
» *ibid.* 20: 2205, 1943.
Normann, E.: Deutsche Ztschr. f. Chir. 212: 166, 1928.
Oberndorfer, S.: München. med. Wchnschr. 75: 683, 1928.
Öwre, A.: Acta chir. Scandinav. 65: Suppl. 13, 1929.
Patey, D. H.: Proc. Roy. Soc. Med. 22: 733, 1929.
Pekelharing, C. A.: Deutsche med. Wchnschr. 18: 1133, 1892.
Philipsborn, E. v.: Folia haemat. 41: 31, 1930.
» Verhandl. d. zweiten Konferenz d. intern. Ges. f. biol. Rhythmusforsch. 1939. = Acta med. Scandinav. Suppl. 108: 192, 1940.
Ponder, C. W.: Lancet 2: 1746, 1908.
Quick, A. J.: Am. J. Physiol. 115: 317, 1936.
» *ibid.* 123: 712, 1938.
Rabinovich, M.: Brit. J. Exper. Path. 8: 343, 1927.
Radvany, A.: Zentralbl. f. Gynäk. 53: 2822, 1929.
Rebaudi, S.: Am. J. Obst. 56: 475, 1907.
Recklinghausen, E. v.: Handbuch d. allg. Pathol. d. Kreisl. u. d. Ernähr. = Deutsche Chirurgie. Lief. 2 & 3, 1883.
Reiss, E.: Der Brechungskoefficient des Blutserums als Indikator für den Eiweissgehalt. Diss. Strassburg 1902.
Rettger, L. J.: Am. J. Physiol. 24: 406, 1909.
Ribbert, H.: Deutsche med. Wchnschr. 38: 1577 & 2270, 1912.
Rich, A. R.: Am. J. Physiol. 43: 549, 1917.
Ritter, A.: Über die Bedeutung des Endothels für die Entstehung der Venenthrombose. Diss. Zürich 1926.
Robertson, H. E.: Am. J. Surg. 26: 15, 1934.
Robertson, T. B.: J. Biol. Chem. 22: 233, 1915.
Scheidegger, W.: Schweiz. med. Wchnschr. 9: 78, 1928.
Schilsky, B.: Ztschr. f. klin. Med. 91: 256, 1921.
Schleussing, H.: Klin. Wchnschr. 8: 2125, 1929.
Schmidt, A.: Zur Blutlehre. Leipzig 1892.

Schmitz, A.: Ztschr. f. physiol. Chem. 222: 155, 1933.
Schulte, H. J.: Ztschr. f. klin. Med. 121: 380, 1932.
Schulz, G.: Deutsches Arch. f. klin. Med. 51: 234, 1893.
Schumacher, P.: Arch. f. Gynäk. 129: 929, 1927.
Seemen, H. v., & Binswanger, H.: Deutsche Ztschr. f. Chir. 209: 157, 1928.
Sköld, E.: On Haemophilia in Sweden and its Treatment by Blood Transfusion. Diss. Stockholm 1944.
Smith, H. P., Ziffren, S. E., Owen, C. A., & Hoffman, G. R.: J. A. M. A. 113: 380, 1939.
Stahl, O.: Deutsche med. Wchnschr. 47: 1550, 1921.
Starlinger, W., & Sametnik, S.: Klin. Wchnschr. 6: 1269, 1927.
Starup, U.: Nord. med. 16: 2876, 1942.
Stewart, J. D.: Ann. Surg. 109: 588, 1939.
Stöhr, W.: Arch. f. klin. Chir. 160: 87, 1930.
Storz, H.: Klin. Wchnschr. 21: 627, 1942.
Strömbeck, J. P.: Zentralbl. f. Chir. 68: 1834, 1941.
Stromberg, H.: Biochem. Ztschr. 37: 177, 1911.
Stuber, B.: Med. Welt 8: 393, 1934.
Sulger, E.: Ergebn. d. Chir. u. Orthop. 24: 326, 1931.
Sulger, E., & Bozsin, Th.: Deutsche Ztschr. f. Chir. 216: 175, 1929.
Syller, R.: Beitr. z. klin. Chir. 145: 322, 1928.
Takaura, G.: Arch. f. klin. Chir. 188: 138, 1937.
Tannenberg, J.: Arch. Path. 23: 307, 1937.
Thordarson, O.: Nord. med. 11: 2079, 1941.
» Undersögelser over prothrombin hos sunde og syge. Diss. Köbenhavn 1941.
Vaquez, H.: Clinique médicale de la Charité. Leçons et mém. 1894.
Vejlens, G.: The Distribution of Leucocytes in the Vascular System. Diss. Uppsala 1938.
Velden, R. v. d.: Arch. f. exper. Path. u. Pharm. 61: 37, 1909.
Vietor, J. A.: Ann. Surg. 82: 193, 1925.
Virchow, R.: Handbuch der speciellen Pathologie und Therapie. Bd 1. Erlangen 1854.
» Gesammelte Abhandlungen zur wissenschaftlichen Medicin. S. 219. Frankfurt a. M. 1856.
Volkert, M.: Studies on the Antithrombin Content of the Blood and its Relation to Heparin. Diss. Köbenhavn 1942.
Vosburgh, C. H., & Richards, A. N.: Am. J. Physiol. 9: 35, 1903.
Walters, W.: Minnesota Med. 10: 25, 1927.
» Surg., Gynec. & Obst. 50: 154, 1930.
Warner, E. D., Brinkhous, K. M., & Smith, H. P.: Am. J. Physiol. 114: 667, 1936.
Westberg, V.: Upsala läkaref. förh. 43: 101, 1937.
Wetterdal, P.: Acta med. Scandinav. 107: 123, 1941.
» Acta obst. et gynec. Scandinav. 23: 707, 1943.
Weymouth, F. W.: Am. J. Physiol. 32: 266, 1913.
Whipple, G. H., & Hurwitz, S. H.: Am. J. Physiol. 13: 136, 1911.

Wilander, O.: Skandinav. Arch. f. Physiol. 81: Suppl. 15, 1939.
Windfeld, P.: Beiträge zur Kenntnis der postoperativen Blutveränderungen. Diss. Köbenhavn 1933.
Wöhlisch, E.: Ergebn. d. Physiol. 28: 443, 1929.
» *ibid.* 43: 174, 1940.
Wöhlisch, E., Diebold, W., & Kiderlen, O.: Pflüger's Arch. f. d. ges. Physiol. 237: 599, 1936.
Wohlgemuth, J.: Biochem. Ztschr. 25: 79, 1910.
» Berliner klin. Wchnschr. 54: 87, 1917.
Yamada, M.: Biochem. Ztschr. 87: 273, 1918.
Zahn, W.: Virchows Arch. f. path. Anat. 62: 81, 1875.
Zopff, G.: Ztschr. f. d. ges. exper. Med. 81: 534, 1932.
Zurhelle, E.: Beitr. z. path. Anat. u. z. allg. Path. 47: 539, 1909—10.

CHANGES OF BLOOD IN CONNECTION WITH THROMBO-EMBOLISM

By G. Bergquist

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Corrections

Page	49	l. 2 fr. above	— add: Several authors consider themselves to have shown that the total protein is decreased after operation.		
»	57	l. 2 & 17 fr. above	— CO factor	*read:*	co-factor
»	62	» 14 fr. below	— above all	»	and above all
»	68	» 4 »	— 35 seconds	»	34 seconds
»	85	» 8 & 10 »	— $^{-2}$	»	^{2}
»	85	» 8 »	— 34 seconds	»	25 seconds
»	93	under Fig. 7	— add: Plotted from the total women material.		
»	93	l. 5 fr. above	— 1.73	*read:*	1.39
»	93	» 7 fr. below	— fall	»	rise
»	96	» 6 »	— normal	»	preoperative
»	98	» 18 fr. above	— by	»	at
»	99	under Fig. 8	— by	»	at
»	125	l. 7 fr. above	— 6.1	»	5.6

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